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The mechanism of antral regulation of gastric secretion: antral pouch studies

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The secretion of acid gastric juice by the fundus of the stomach ceases when the pH of the antrum falls below 1.5. Since this response is also observed in acid-secreting fundus mucosa which is denervated and isolated (Heidenhain pouch), it is presumed that the effect is mediated by humoral factors of antral origin. Beyond this point of explanation, however, experimental evidence becomes conflicting and even contradictory.

Oberhelman and associates[5] noted that when acid-secreting gastric mucosa was left in continuity with the gastric antrum, less gastric secretion occurred than when it was removed from continuity. Woodward and his group[8] then demonstrated that the gastric secretory response to crude liver extract at pH 6.0, placed in an isolated, innervated antral pouch, was blocked by prior introduction of 0.1 N hydrochloric acid. These and other investigations[4] have given rise to the hypothesis that gastric secretion is checked by paralysis of the mechanisms responsible for the release of the antral stimulatory hormone, gastrin.

Harrison and colleagues,[1] however, observed that the gastric secretory response of a dog in which the distal half of the antrum had been transplanted into the colon was increased by removal of the remaining (proximal) half of the antrum. State[6, 7] then showed that greater protection against histamine-induced ulceration in the dog could be achieved by leaving the antrum in continuity than by removing it entirely. Additional observations, notably by Jordan and Sand,[3] have given impetus to the view that regulation of gastric secretion is accomplished by active inhibition of parietal cell activity, presumably by means of a humoral agent elaborated by the antrum in response to a falling pH.

The role of the vagus nerve has not been clarified within the framework of either hypothesis. However, there is general agree-

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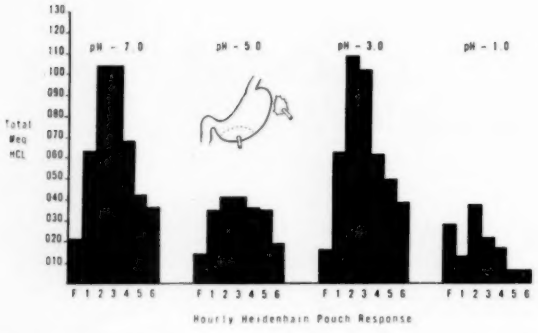

Fig. 1. Hourly secretory response to standard test meal according to pH in intact antral pouch.

ment that a denervated antrum does not lose its ability to stimulate gastric secretion.[2]

Our experiments were carried out in an effort to clarify some of these divergent views.

MATERIALS AND METHODS

Six adult mongrel dogs which weighed from 15 to 20 kilograms each were used for the study. Each dog underwent construction of a Heidenhain pouch which was cannulated to the exterior by a flanged plastic cannula. At a second stage, a vagally intact, lumen-isolated pouch, cannulated to the exterior, was prepared from approximately one-half of the gastric antrum by the mucosal bridge technique. The construction of these pouches was carefully tailored so that the other half of the antrum remained in normal continuity, free of pyloric outlet obstruction.

After a 4-week convalescence, each dog was studied. Basal secretion was collected from the Heidenhain pouch for 1 hour. The animal was given a standard test meal (one-half can of Pard dog food), and collections made from the Heidenhain pouch hourly for 6 hours. As soon as the meal had been ingested, the antral pouch was gently perfused with normal saline solution buffered either to pH 7.0, 5.0, 3.0, or 1.0 with 0.1 N hydrochloric acid. The perfusion was continued throughout the 6-hour collection period. No less than 30 daily observations were made at each perfusion pH. A random assignment technique was used so that all studies with similar perfusion pH were not performed consecutively. One milliliter of each hourly Heidenhain pouch collection was titrated with 0.1 N sodium hydroxide with Toepfer's reagent and expressed as milliequivalents of hydrochloric acid by multiplication by the volume collected.

At the conclusion of the study 3 dogs were again operated upon and the antral pouch vagotomized by separation from the stomach. After a convalescence of 1 month the entire study was repeated.

RESULTS

The general pattern of the secretory response of the Heidenhain pouch to the test meal was remarkably consistent among all dogs. Basal secretion was generally low, with peak response occurring during the second or third hour after the meal. There was a relatively rapid fall in secretion to approximately the basal level in the sixth hour. Although this pattern was consistent among all dogs there was considerable variation in the quantitative response according to the pH in the antral pouch.

Fig. 1 illustrates these differences. Note the excellent secretory response of the Heidenhain pouch to the test meal which occurred when the pH of the antral pouch was high. Note also the general tendency of the secretory response to parallel the change in antral pH.

After the antral pouch had been vagotomized by separation from the stomach, several interesting changes took place. These

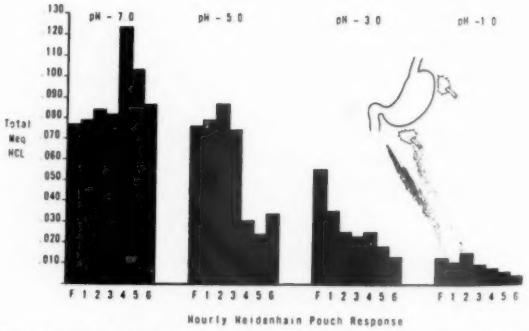

Fig. 2. Hourly secretory response to standard test meal according to pH in isolated antral pouch.

are apparent in Fig. 2. Basal secretion was significantly increased, and the tendency for the quantitative secretory response to parallel the pH in the antral pouch was further enhanced. Note particularly that the secretory response was only half as great in dogs with a vagotomized antrum at the low pH levels.

DISCUSSION

Although subject to several interpretations, the results observed warrant comment. Since half of the antrum remained undisturbed (that is, vagally intact and in continuity) throughout all of the experiments, its contribution to the secretory response after a test meal was presumably constant. Therefore, the quantitative differences in the secretory responses must have been a function of the pH in the isolated antral pouch. Since these responses were greatest when its pH was highest it would seem that 2 mechanisms were operating, (1) the antral pouch was the source of additional gastrin when its pH was high and, (2) when its pH was low it was capable of inhibiting the secretory response to stimulation of the intact antrum. The first mechanism is entirely reasonable. Woodward and his group[8] have shown conclusively that the release of gastrin by the antrum is greatly enhanced if an elevated antral pH is induced prior to its distention. The second explanation, however, needs clarification.

If the thesis is correct that a low antral pH blocks the release of gastrin, then, in an experiment such as the one described, the poor secretory response to a test meal associated with perfusion of the vagally intact but lumen-isolated antral pouch with solutions at low pH would be expected. On the other hand, if this were the only explanation, subsequent surgical isolation of the antral pouch so that local "neuroreflex" arcs are interrupted should yield a secretory response equal to, or greater than, that observed while the reflexes were intact. *We observed the opposite.* The secretory response at low pH perfusions of the separated pouch were approximately one-half those observed before its attachments were severed. This observation is difficult to interpret without the thesis that active inhibition, humoral in nature, was taking place.

CONCLUSIONS

1. The secretory response of a Heidenhain pouch to a test meal was determined in dogs with one-half of the antrum in normal continuity and the vagus intact.

2. Simultaneous perfusion of the remaining half of the antrum, lumen-isolated but vagally intact, enhanced the secretory response when solutions of high pH were used, but inhibited the response when solutions of low pH were used.

3. Repeat studies after physical separation of the antral pouch showed further increase in the Heidenhain pouch response to a test meal when the antral pouch was perfused with solutions at high pH but half as much response when the pouch was perfused with solutions at a pH of 1.0.

4. Since paralysis of the gastrin-release mechanism in the intact antrum was impossible under the conditions of this experiment, it appears that active inhibition may have been achieved.

REFERENCES

1. Harrison, R. C., Lakey, W. H., and Hyde, H. A.: The production of an acid inhibitor by the gastric antrum, Ann. Surg. **144:** 441, 1956.
2. Jones, T. W., Stevenson, J. K., Jesseph, J. E., Nyhus, L. M., and Harkins, H. N.: Heidenhain pouch secretory response to transplantation of the innervated and denervated antral pouch to the transverse colon, Ann. Surg. **147:** 13, 1958.
3. Jordan, P. H., and Sand, B. F.: Antral inhibition of gastric secretion, Proc. Soc. Exper. Biol. & Med. **94:** 471, 1957.
4. Longhi, E. H., Greenlee, H. B., Bravo, J. L., Guerrero, J. D., and Dragstedt, L. R.: Question of an inhibitory hormone from the gastric antrum, Am. J. Physiol. **191:** 64, 1957.
5. Oberhelman, H. A., Jr., Woodward, E. R., Zubiran, J. M., and Dragstedt, L. R.: Physiology of the gastric antrum, Am. J. Physiol. **169:** 738, 1952.
6. State, D.: The role of the gastric antrum in experimental ulceration and regulation of

gastric secretion, Gastroenterology **38:** 15, 1959.

7. State, D., and Morgenstern, L.: The inhibitory role of the pyloric antrum in the cephalic phase of gastric acid secretion in dogs, Surg. Gynec. and Obst. **106:** 545, 1958.
8. Woodward, E. R., Lyons, E. S., Landor, J., and Dragstedt, L. R.: The physiology of the gastric antrum: experimental studies on isolated antrum pouches in dogs, Gastroenterology **27:** 766, 1954.

Malignant lesions of the stomach in Hawaii: experiences with 350 patients

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In several previous publications it has been pointed out that cancer of the stomach occurs with comparatively greater frequency in Hawaii than on the mainland of the United States because of the high percentage of people of Japanese ancestry in the Hawaiian population.[17, 23, 25, 26] To date, no one has offered a satisfactory explanation for the susceptibility of the Japanese to gastric cancer. Statistics from the Board of Health and our own experience at the Straub Clinic, as shown in Tables I through IV, bear out this conclusion.

In this study, our observations are based on 350 patients with gastric malignancy operated upon by members of the Surgery Department of the Straub Clinic since Jan. 1, 1935; 283 of these were operated upon 5 or more years prior to this publication. A follow-up has been obtained on 100 per cent of the 283 patients and on all but 1 of the total 350 patients. Three hundred two of the patients are known to be dead. Of the 283 patients with follow-up for 5 years or more, 28 are still living and 37 lived for at least 5 years (Table V).

CURATIVE RESECTIONS

In the total 350 cases, there were 55 curative resections. A curative resection is understood by us to mean one in which no gross extension of the malignancy is found outside of the stomach at the time of operation and in which the operative findings are subsequently confirmed by careful microscopic examination of all tissue removed. Forty-seven of the curative resections were performed on the 283 patients followed for 5 years. Twenty-seven, or 57.4 per cent, lived for 5 years or longer. Eighteen of the remaining 20 who survived operation lived for an average of 1.1 years. There were 103 of the 283 resections that were not curative, that is, metastases were present. The average length of life in these cases was 1.6 years. Just why the 18 patients who had curative resections failed to live as long or even longer than those in whom metastases were demonstrated is not at once apparent. Undoubtedly, spread of the disease had already occurred beyond the regional lymph nodes and other factors, such as virulence of the disease and host resistance, must have come into play.

It has been shown by many that the best results in the surgical treatment of carcinoma of the stomach are obtained when the lesion cannot be demonstrated to have spread beyond the stomach. In 1940, Pack and Livingston[21] found that approximately 50 per cent of such patients lived for 5 years. Ransom[22] of the University of Michigan made the same observation. Blalock and Ochsner[4] found a 5-year survival of 63 per cent of 19 patients with distal subtotal gastrectomies which were classified as curative resections. Welch and Wilkins[31] found that the 5-year survival rate of patients without nodal metastases who recovered from the operation in 1937 was 48 per cent and in 1951, 51 per cent. Marshall,[14] in studying 1,666 patients with gastric carcinomas who underwent operation, reported that 39.1 per cent underwent curative resection and that the 5-year

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Table I. *Racial make-up of population in Hawaii, 1950 census*

	No.	*%*
Japanese	184,611	36.9
Caucasian	114,793	22.9
Hawaiian or part Hawaiian	86,091	17.3
Filipino	61,071	12.3
Chinese	32,376	6.4
Other	20,852	4.2
Totals	499,794	100.0

Table II. *Cancer death rates per 100,000 persons for continental United States and Hawaii*

	U.S.A. 1955	*1954-1956* Hawaii (crude)*	*1954-1956† Hawaii (adjusted)*
Cancer, all sites	146.5	102.9	159.1
Cancer of stomach	13.5	23.7	36.0
Cancer of lung	16.3	13.7	21.4

*Average population.
†Adjusted for age to total United States population estimated for 1955.

survival rate for them in 1954 was 39.5 per cent.

Of the 37 patients who lived for 5 years after resection, 10 had regional lymph node metastases, but in none of these had the malignancy spread beyond the regional lymph nodes. Three of the 10 patients with metastases to the lymph nodes who lived more than 5 years have died. The average length of life after operation was 6.7 years. Six of the patients without nodal involvement who lived more than 5 years have died and the average length of life was 8 years.

HISTOLOGIC GRADING

We have not attempted to grade the degree of malignancy of all the lesions resected, for we have long since lost faith in this method of estimating prognoses. Too many of the patients in whom lesions were considered by their microscopic appearance to be relatively benign have died in a short period of time of metastases, while several of the patients have survived for many years even though the microscopic findings indicated a high degree of malignancy. This is exemplified by Figs. 1 and 2, which show the radiologic and microscopic appearance of the resected specimens from patients living and well 18½ and 17 years, respectively, after operation. The microscopic picture of each indicates a high degree of malignancy. Marshall[14] recently stated, "I thought we had gained something from histologic study but after reviewing the pathologic findings in our series of cases, we found nothing in the microscopic anatomy that helped us to determine the survival rate."

We are aware that other observers have placed considerable reliance on the grading of such malignancies, but it has been our experience that the presence or absence of spread beyond the stomach is of far greater significance, and such spread does not correlate well with the histologic grade. Berkson and associates[3] concluded that with gastric carcinoma classified as Broder's grade I and II, one might expect a 15-year survival rate of 33.3 per cent, with grade III 18.8 per cent, and with grade IV 12.7 per cent. Harvey and his group[8] also came to the conclusion that the pathologic type of malignancy was of significance in the estimation of the patient's life expectancy.

DO BENIGN GASTRIC ULCERS BECOME MALIGNANT?

The question of whether or not benign ulcers of the stomach ever undergo malignant degeneration and, if so, how frequently has long been debated. Most writers on this subject, without any very convincing evidence to confirm their conclusions, are of the opinion that this may occur, but rarely—perhaps in 2 to 5 or 6 per cent of the cases. It was formerly believed that such a transformation occurred very frequently. Wilson and MacCarty[32] came to the conclusion that such degeneration had occurred in 71 per cent of the cases they had studied. Finster[6] stated that malignant degeneration of a benign ulcer occurred so frequently that re-

section of a gastric ulcer could be regarded as an early operation for cancer of the stomach. Marshall[15] studied a group of 411 consecutive gastric ulcer patients in whom surgical resection had been performed. The incidence of malignant ulcer in this group was found to be 15.8 per cent. He decided that "there is insufficient histologic evidence to warrant a conclusion that gastric ulcers present a high potential origin for malignancy, but that unquestionably malignant degeneration does occur in a chronic ulcer but probably in no more than five to six per cent." Our pathologists have ventured an opinion that in a number of our resected specimens, the malignancy originated in a chronic benign gastric ulcer. So far as I have been able to determine, there are no unrefutable criteria by which such a conclusion can be accurately reached, although such observations seem to be of some significance. Since chronic irritation has long been recognized as a contributing factor in the development of malignancy in other parts of the body, it would be strange indeed if a chronic gastric ulcer, constantly irritated by acid gastric content and recurring peristaltic activity, did not react in a similar fashion, especially in an organ so prone to the development of malignancy.

In 60 of the 283 patients with gastric cancer, the history definitely stated that epigastric distress and discomfort had been experienced for from one to many years. A number had had repeated roentgenographic examination over the years without any evidence of an organic lesion being demonstrated. Fig. 3 illustrates this point. Some, I am sorry to say, had been treated medically for a demonstrated ulcer for months or even years before operation, and surgical treatment was then usually too late to be of much value. Fig. 4 illustrates such a case. In the 60 patients mentioned, 18 had had symptoms for approximately 1 year, 11 for from 2 to 3 years, 10 for from 3 to 5 years, and 21 for from 5 to 20 years. This observation, of course, proves nothing, but it seems to be significant circumstantial evidence that there may have been an exciting factor, probably an ulcer, at work for a considerable period of time before malignancy developed.

Another suggestive observation made by many is that the longer supposedly benign

Table III. *Gastric cancer in Hawaii—morbidity study of 4,191 patients with accepted malignancies, 1944-1954**

	No.	*%*
No. of cancers in males, all races	2,269	
Gastric cancer in males, all races	498	21.0
No. of cancers in females, all races	1,922	
Gastric cancer in females, all races	212	11.0
Total cancers, all races, both sexes	4,191	
Gastric cancer, all races, both sexes	710	16.9

*From cancer morbidity study, Hawaii, 1944-1954, Bureau of Cancer Control, Hawaii, 1958, p. 76.

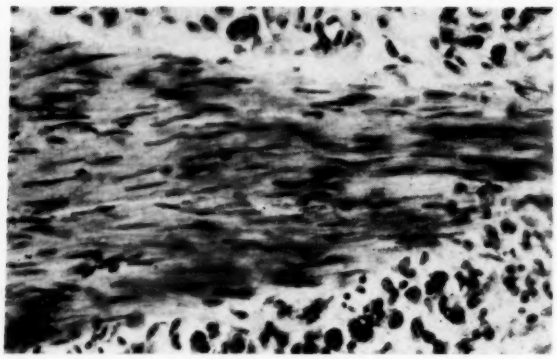
Table IV. *Race and sex make-up of 350 patients with gastric carcinomas operated upon by surgical department, Straub Clinic, 1935-1959*

Race	*Males*	*(%)*	*Females*	*(%)*	*Both*	*(%)*
Japanese	169	48.3	67	19.1	236	67.4
Caucasian	51	14.5	10	2.9	61	17.4
Hawaiian or part Hawaiian	10	2.9	8	2.3	18	5.2
Chinese	8	2.3	8	2.3	16	4.6
Filipino	5	1.4	1	0.3	6	1.7
Other	10	2.9	3	0.8	13	3.7
Totals	253	72.3	97	27.7	350	100.0

Table V. *Follow-up results in 350 patients with gastric cancer*

	No.	*%*
Patients with follow-up	349	
Patients followed 5 years or longer	283	
Patients alive 5 years or longer	37	13
Number in latter group with regional node involvement, none extensive	9	
Still alive	28	

A



B

Fig. 1. *A,* Roentgenogram which shows carcinoma of the stomach in a part-Hawaiian woman, 35 years of age, who is living and well 18½ years after undergoing gastric resection. At operation no evidence of spread beyond the stomach had been found. *B,* Photomicrograph which shows diffuse infiltration of the muscle layers by anaplastic carcinoma.

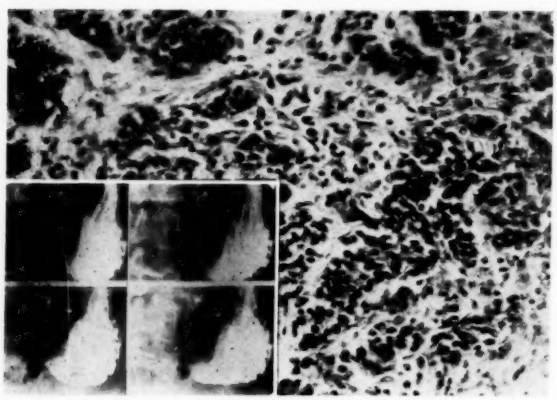


Fig. 2. Small penetrating ulcer in the lesser curvature of stomach, which was treated medically for 5 months, in a Caucasian man, age 42. There are no lymph node metastases. The photomicrograph shows anaplastic carcinoma. The man is living and well 17 years after gastric resection.

ulcers are treated medically and the more frequently they recur, the more likelihood there is that the lesion will ultimately prove to be malignant.

Perhaps the recently developed method of Wangensteen and his associates of demonstrating the uptake of radioactive phosphorus by malignant lesions of the stomach by the use of an intragastric radiosensitive balloon may help in following the course of benign gastric ulcers.

AGE

The age incidence in this series is of some interest. While the average age for the 350 patients was 57 years, there were 5 patients in their twenties and 5 in their eighties. Of the former, 4 were women, aged 27, 28, 29, and 29 (Korean, part Hawaiian, Puerto Rican, and Chinese, respectively), and 1 was a Japanese man, age 22. They all underwent gastric resection and the longest survival was 1 year and 9 months. Four of the octogenarians were Japanese. Two were men, age 81 and 83, and 2 were women, age 80 and 81. The other was a Caucasian man of 80. Four of these patients underwent gastric resection. Two are living, 2 died in the early postoperative period, and the other survived exploratory operation and lived a few months. Tamura and Curtiss,[29] in a recent study of carcinomas of the stomach occurring in young adults treated in the 3 major general hospitals in Honolulu over a 10-year period (1948 to 1958), found 28 patients under 36 years of age. They also found a predominance of such cancers in women, the ratio being 1 man to 2.5 women.

SITE OF THE LESION

It was also of interest to determine the site of the malignancy, though it is recognized that some of the lesions were widespread, particularly the linitis plastica type. In some instances, when only an exploratory operation was performed, this point was not clearly stated and undoubtedly was not easy to determine. Both the roentgenographic report and the operative findings were used for this determination. In 175 patients the

lesion was in the pyloric area and at least 53 of these had varying degrees of pyloric obstruction. In 47 the lesions occurred in the proximal portion of the stomach and in 128 the lesions involved the midportion or the entire stomach, or the sites were not clearly stated. Of the 37 patients who lived for at least 5 years postoperatively, the lesion was in the distal portion of the stomach in 29, in the lesser curvature in 6, in the greater curvature in 1, and in the midportion of the stomach in 1. No patient lived for as long as 5 years when the primary site of the lesion was in the proximal part of the stomach.

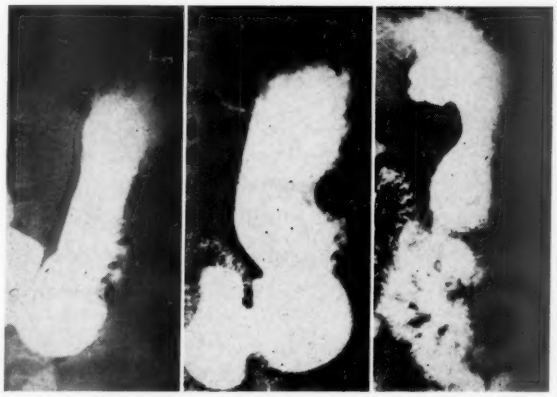
TYPE OF OPERATION

Of the 350 patients who underwent operation, 175 had a gastric resection, 46 total gastrectomy, 28 gastroenterostomy, and 101 had only an exploratory operation. In a few of the latter in our earlier experience, we added a gastrostomy. For all practical purposes, this has been discontinued because it was not found to add to the patient's comfort. We believe, along with many others, that resection is frequently indicated, particularly in the presence of obstruction and hemorrhage, even though it is quite evident that it is only a palliative procedure. We have been agreeably surprised at how long some individuals have lived in comfort after this procedure.

In 100 cases, it was recorded that the patient had a palpable epigastric mass. Ninety-six of these patients have died and only 2 survived for as long as 5 years. In Marshall's series of cases,[14] a palpable mass was present in 30 per cent and about one-third of these patients could be operated upon but none survived 5 years.

MALIGNANCIES OTHER THAN CARCINOMAS

In this series of 350 cases, there were 5 lymphosarcomas and 1 leiomyosarcoma. Three of the lymphosarcomas occurred in men. Two had only exploratory operation, 1 lived 1 month, and 1 lived 8 months. The third lived 8 months after gastric resection. One woman had a resection and lived 5 months, while the other had a total gastrectomy and lived 11½ months. The leiomyosarcoma occurred in a man who lived 3 years after resection and died of unrelated causes.



A *B* *C*

Fig. 3. Japanese woman, age 59, had intermittent epigastric distress for 10 years. Gastrointestinal x-ray studies made from 1951 to 1957 (*A* and *B*), appear to be negative. Roentgenogram made in June, 1960 (*C*), shows a large penetrating ulcer in the proximal lesser curvature. At operation the lesion was found to be a carcinoma penetrating into the tail of the pancreas. A total gastrectomy including resection of one-third of the pancreas was carried out.

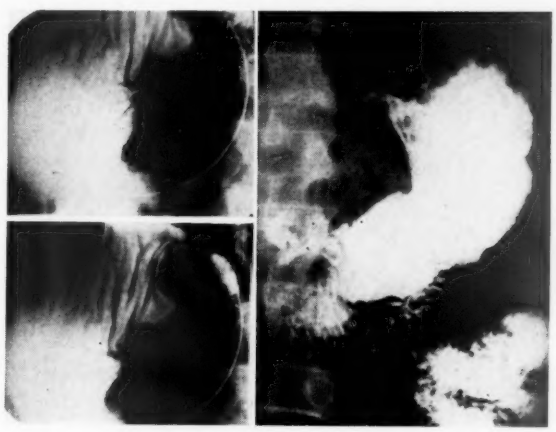


Fig. 4. A 39-year-old Caucasian man was treated medically for ulcer for 2 years. The 2 roentgenograms on the left, taken in October, 1956, were interpreted by the radiologist as showing an ulcer of the stomach, probably benign. Roentgenogram at right, taken 5 months later, was thought to show ulcerating carcinoma in the antrum. Exploration showed free abdominal fluid and extensive metastases. Patient died soon afterward.

Giberson and colleagues[7] stated that 98 per cent of malignant neoplasms of the stomach are adenocarcinoma and that lymphosarcomas, leiomyosarcomas, fibrosarcomas, and angiosarcomas make up the remaining 2 per cent. Leiomyosarcomas, they found, comprised about 25 per cent of the sarcoma group. Mosley,[19] in reviewing this subject, found that leiomyosarcoma occurred once in about 1,000 malignancies of the stomach. The usual findings in this condition are an epigastric mass associated with pain, hemorrhage, and anemia. Such neoplasms are slow to metastasize and radical removal is attended by a high percentage of 5-year survivals.

NUMBER OF RESECTIONS

Combining resections and total gastrectomies, there were 233 cases in which either an attempt at cure was made or the bulk of the tumor was removed for palliation. Many of the resections were recognized at the time as being palliative only. This gave an over-all resection rate of 66.5 per cent. In the first 175 cases (Jan. 1, 1935, to Jan. 1, 1952), there were 107 resections (61.1 per cent), and in the last 175 cases, 126 resections (72 per cent).

MORTALITY RATE

The over-all mortality rate of the 350 patients who underwent operation was 10 per cent when only patients who died during the first 3 postoperative weeks are counted. In the first 175 cases there were 19 deaths (10.8 per cent) and in the last 175 cases there were 16 deaths (9.14 per cent). Included in these figures were 46 patients who underwent total gastrectomies with a mortality rate of 17.1 per cent. There were 107 resections in the first 175 cases with a mortality rate of 12.1 per cent and 126 resections in the second series with a rate of 11.1 per cent. The over-all survival rate of the 283 patients followed for 5 years was 13 per cent. Of the 37 who survived for 5 years, 28 are still living. Eighteen are men, 16 of whom are Japanese, and 10 are women, 8 of whom are Japanese.

The mortality rate after various types of operations used in the treatment of carcinoma of the stomach varies considerably in most people's experience. In addition to the skill and experience of the surgeon and the assistance he may have, which includes the anesthesiologist, his surgical team, and physical equipment, there are many other factors to be considered. All things being equal, if only good-risk patients are selected for operation, the mortality rate should be considerably less than for the surgeon who believes that under such circumstances all patients should be given the opportunity of surgical investigation. Again, if resections are performed only when cure might be possible, the mortality rate should be less than when palliative resections are believed justifiable. Radical resection of the stomach, which includes removal of a portion of the various contiguous organs, will result in higher mortality rate than a more conservative resection.

It is our conviction that all individuals strongly suspected of having or diagnosed as having a malignancy of the stomach are entitled to exploratory operation unless there are definite contraindications. Such contraindications should be absolute, for under such circumstances everything is to be gained and relatively little to be lost. Three examples illustrate these points.

CASE REPORTS

Case 1. A man of 76 presented himself with symptoms indicative of gastric malignancy. Roentgenograms showed a filling defect in the cardiac end of the stomach and there was a palpable nodule above the left clavicle. This was removed in the office and was found to be a lymph node filled with metastatic carcinoma. Since the patient had no obstructive symptoms, further surgical treatment was not advised. This patient died several months later and autopsy showed carcinoma of the stomach with extensive metastases to the liver and other viscera. The patient meanwhile had had very little discomfort.

Case 2. An 82-year-old man with epigastric distress of several months' duration had a palpable epigastric mass and the report on roentgenographic examination of the stomach stated, "There is a hard, tender mass adjacent to or involving the stomach which is probably a linitis plastica type of infiltration of the stomach wall,

Fig. 5. Roentgenograms of patient diagnosed preoperatively as having gastric carcinoma. Resected specimen showed a benign ulcer.

with partial destruction of the mucosa and diffuse carcinomatous infiltration." The family was advised that because of the patient's age and our physical and roentgenographic findings, in all probability the patient had an advanced carcinoma of the stomach and that operation, if he survived the ordeal, would be of little or no value. The patient himself wished to undergo exploratory operation because he knew of an elderly man with similar findings in whom a benign ulcer had been found at operation. We were somewhat encouraged to proceed, because an attempt at gallbladder visualization had been unsuccessful. At operation, a ruptured gall bladder was found containing stones. The epigastric mass was inflammatory and had caused the changes in the gastric wall described by the roentgenologist. Results of exploration of the interior of the stomach were negative. After cholecystectomy, the patient made an uneventful recovery.

Case 3. A Japanese woman, age 63, with epigastric distress, weakness, and loss of weight of several months' duration, was diagnosed preoperatively as having a carcinoma of the stomach. The roentgenologist's report read, "Study of the gastrointestinal tract shows a large ulcerating and infiltrating malignancy of the stomach measuring 6 by 2 cm. in diameter, penetrating the middle third of the lesser curvature. There is adjacent destruction of the mucosa and infiltrating and polypoid lesions of the antrum. Resectability is doubtful." Fig. 5 shows a roentgenogram of this patient.

On Jan. 9, 1959, the lesion was widely resected to include the entire omentum, spleen, and surrounding structures because we thought without doubt that we were dealing with an advanced gastric carcinoma. To our surprise, the lesion proved to be a benign gastric ulcer and the patient made an uneventful postoperative recovery.

On other occasions we have been morally sure that we were dealing with an advanced malignancy of the stomach, only to be happily surprised to find that the patient had a perforated, but walled-off, benign ulcer. On other occasions lesions which had appeared preoperatively to be inoperable carcinoma were found to be easily resectable on exploration. One should always remember that the abdominal wall has a habit of concealing the true nature of what lies beneath.

SURVIVAL RATE IN CANCER OF THE STOMACH

The over-all survival rate of people suffering with cancer of the stomach has improved over the years but, as physicians, we have little to be proud of in comparison to what has been achieved in other fields of medicine. Results could improve several hundred per cent and there still would be much to be accomplished. While it is true that 100 per cent of patients with untreated cancers of the stomach will ultimately die of their disease, even the best postoperative statistics that I have been able to find, originating from the Mayo Clinic,[3] leave much to be desired. Of all patients seen with this disease, 14 per cent survived for 5 years. From 1907 to 1916, the 5-year survival rate at this clinic had been 5 per cent. At the Ochsner Clinic,[4] 80 per cent of such patients were considered to have inoperable lesions at the time of diagnosis. Ransom[22] of the University of Michigan reported that the number of 5-year survivals after definitive surgical treatment based on the total number of patients seen, 1,264, was 7.8 per cent. Beale and Hill[2] found that 9.3 per cent of 505 patients admitted to the New York Hospital survived for 5 years or longer. Wangensteen[30] stated in 1956, "In our own experience of patients with gastric cancer exhibiting no lymph node involvement who have had gastric resection for cure, there

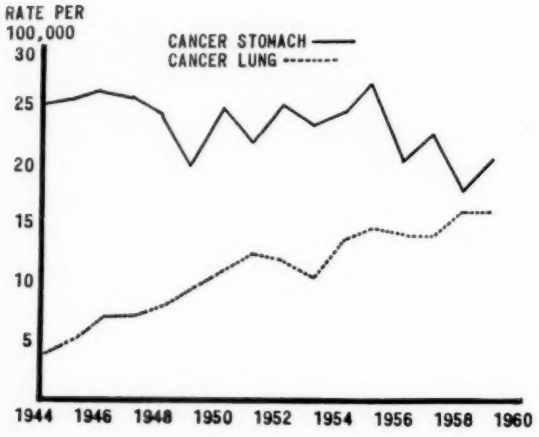


Fig. 6. Cancer rates in Hawaii.

are more than 50 per cent five-year survivors. For the year 1944, in the lymph node negative gastric resection group (operative mortality excluded), there were 64 per cent alive and well five years later. For all patients with gastric cancer seen at the University Hospital during the period from 1946 to 1949, the survival rate was 12.5 per cent." In our own statistics, there was a 5-year survival rate of 13 per cent of all patients who underwent operation. There were very few patients in whom a diagnosis of cancer of the stomach was made in whom surgical exploration was not carried out.

DISCUSSION

It is evident from our statistics from the Straub Clinic, as well as from data from all other such studies, that the treatment of cancer of the stomach at the present time leaves a great deal to be desired. Rarely do patients suffering from this disease consult their physician sufficiently early to enable the surgeon to perform a curative resection, and even though the patient does consult his physician early, the diagnosis is not always correct. By the time a clinical diagnosis of cancer of the stomach is made, it is usually too late to hope for more than palliative assistance, if any help at all is possible. We are convinced that individuals in whom a definite diagnosis cannot be made but in whom the indications are those of cancer of the stomach, should undergo exploratory operation. We have encountered this difficulty, particularly in patients with hypertrophic gastritis. In our opinion[26] it is not too big a price to pay—even though some results of exploration are negative—if by this method we can perform a curative resection in a patient whose disease would otherwise reach a stage of inoperability. It has been stated by many observers that cancer of the stomach is on the decrease. Marshall[14] has recently written, "Cancer of the stomach has decreased over a ten-year period. In 1948 cancer of the stomach accounted for 13.3 per cent of all deaths from cancer; in 1957 that figure was 8.5 per cent. This decrease in deaths from cancer of the stomach is surprising since life expectancy has increased to about 70 years and according to the Bureau of Vital Statistics, the number of deaths from cancer in general has increased." That cancer of the lung is markedly on the increase almost everywhere is well known. Fig. 6 indicates that these observations on cancer of the stomach and of the lung are also applicable to Hawaii.

The statement has been made by MacDonald[12] and others that the early diagnosis of carcinoma of the stomach is not a factor in improving end results of gastric operations. So far as I am concerned, this is an entirely fallacious conclusion, and I am convinced that such teaching, if widely accepted, will do much to undermine the advances that have been made in combating cancer wherever encountered. While it is true that most statistics have shown that the longer the patient's symptoms have existed, the better the prognosis, this is by no means an indication that early diagnosis and surgical intervention are not of great importance. Swynnerton and Truelove,[28] speaking of cancer of the stomach, stated, "an outstanding finding of the present study is the marked prognostic significance of length of history. Among patients with surgical resection, the greater the length of history, the better the prognosis." Our own statistics are in agreement with such conclusions. However, regardless of the type of lesion and whatever its biologic characteristics may be, no one,

I am sure, would dispute the fact that the longer a cancer is left in place, the greater the probability of metastases, and this possibility, in our experience and in the experience of nearly everyone, is the most important factor in the determination of prognosis.

It has been claimed by Welch and Wilkins[31] and others that the employment of more radical surgery in the treatment of cancer of the stomach has resulted in more 5-year survivals, and perhaps more cures. It has not been our experience that superradical operations for this condition have resulted in sufficient improvement in end results, considering increased mortality rate and morbidity, for us to be very impressed about such an approach to this problem. Perhaps this is largely due to our being less expert in carrying out the technical aspects of such radical operations. In a recent discussion on the employment of superradical operations in the treatment of cancer of the breast with Owen Wangensteen, he stated that his results had not justified his continuing to advocate this type of operation for this condition. I am fully convinced that this also holds true in the treatment of cancer of the stomach, especially when one considers the rank and file of surgeons who are called upon to perform such operations. Our own experience with total gastrectomy, while limited, has been disappointing. Of the 46 patients with total gastrectomies, 2 are living (1 for 6 years and 8 months) without evidence of recurrence. The other underwent operation recently. Only one other patient lived for as long as 5 years. When the malignant process has reached such proportions that the gross evidence of the disease can be removed only by this method, the process in most cases has already spread beyond the reaches of the area resected and hope of eradicating the disease, in our experience at least, has been largely a figment of the imagination.

During the time in which we have operated upon the 350 patients with carcinoma of the stomach at the Straub Clinic, we have resected 413 ulcerating lesions of the stomach which proved to be benign. We have been asked what percentage of the lesions which we had thought preoperatively to be benign proved to be malignant, and what percentage of the ulcers we had thought malignant proved to be benign. We are unable to answer this question. We know of no accurate method of determining preoperatively in many cases which ulcerating lesion is benign and which is malignant. Therefore, we consider all ulcerating lesions of the stomach to be malignant until careful microscopic examination of the ulcer and other removed tissue has been made and has shown no evidence of malignancy. We do know that a number of ulcers have been treated medically for varying periods of time that later were proved to be malignant. We know that some of the patients responded favorably to such treatment for a period of time. We know that the most frequent favorable results in the treatment of cancer of the stomach have followed removal of ulcers that clinically might be expected to be benign, and that rarely in our hands does a patient survive for 5 years when preoperatively the lesion has the characteristics of malignancy and is so proved by microscopic examination. From reviewing the experience of others as published in current literature, we are convinced that the medical treatment of ulcerating lesions of the stomach in the majority of instances is unsatisfactory even though the lesion is benign.[1, 5, 9, 13, 16, 20, 28]

We have convinced ourselves from our own experience that results from resection of the stomach for benign ulcers give satisfaction in a very high percentage of cases. The mortality rate is very low, complications are few, and patients rarely are displeased with end results.

In our present state of knowledge, it seems that no one can hope to improve very materially the present inadequate approach to the problem of cancer of the stomach by surgical means. Whatever improvement is accomplished will not be gained by extending the scope of the operation, but it will be achieved by bringing patients to operation by earlier diagnosis, when the lesion is still

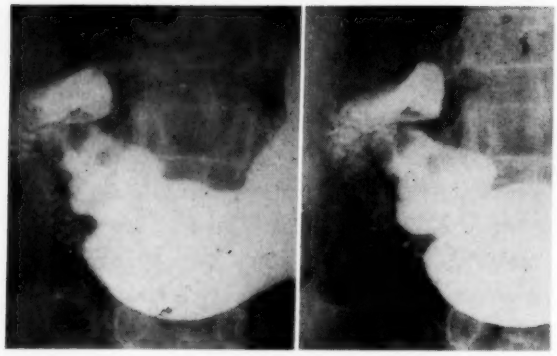


Fig. 7. Roentgenograms of a Japanese woman, age 53, show constant filling defect in pyloric region due to a polyp. The lesion was removed locally in 1943 and the patient has had no trouble since.

Fig. 8. Roentgenogram of a Caucasian woman, of polyps. Gastric resection was performed in age 65, shows multiple filling defects as a result 1952 and the patient has been well since.

confined to the stomach, and by removing lesions that are known to be precursors of malignancy or that may in their incipiency be impossible to differentiate from early malignancy. Polyps of the stomach are recognized as being a source from which cancer develops. While occurring relatively infrequently, they should be removed whenever diagnosed. If the lesions are localized and grossly benign, and this is substantiated by microscopic examination, local excision may suffice. Fig. 7 shows the roentgenogram of a polyp removed by local excision. The patient has remained well for 17 years, and frequent x-ray studies have shown no recurrence of polyps. Fig. 8 shows multiple polyps removed by partial gastric resection. This patient has also remained free of symptoms with negative roentgenographic studies for 8 years.

McManus and Sommers[18] studied 45 patients with gastric polyps and showed that 13.3 per cent developed gastric cancer before they died. In patients with pernicious anemia, cancer of the stomach has long been recognized as a frequent complication, and for this reason, frequent roentgenographic examination of the stomach is advisable. Hitchcock and associates[10] found that patients with pernicious anemia had gastric carcinoma 21.9 times more frequently than an equal segment of normal population. Kaplan and Rigler[11] emphasized the high frequency of association of gastric polyps, 7 per cent, and gastric carcinoma 12.3 per cent, in patients who die with pernicious anemia. The latter was 3 times the expected autopsy incidence of gastric carcinoma for the same age group.

The most discouraging fact remains that the vast majority of malignancies of the stomach progress to a stage of incurability before producing symptoms and signs of sufficient severity to induce the patient to seek adequate medical attention. Even though such patients do seek our council sufficiently early, we too frequently are not able or fail to make a correct diagnosis.

CONCLUSIONS

From this study and from other similar investigations, one can only conclude that the present-day treatment of cancer of the stomach is most unsatisfactory. The most favorable prognostic evidence for long survival after operation is the absence of any demonstrable evidence, either grossly or by microscopic examination, of spread of malignancy beyond the stomach. If the immediate

regional lymph nodes are involved, the chances of 5-year survival are diminished. When extension beyond this area has occurred, 5-year survivals are very exceptional. It has been our experience that total gastrectomy and radical subtotal gastrectomy, which include removal of the spleen, omentum, and at times a portion of the pancreas or transverse colon, or the left lobe of the liver and other contiguous structures, rarely contribute to long survival of the individual. Superradical operations under these circumstances, in our experience, are seldom rewarding and are rarely indicated. Earlier diagnosis by methods which we hope to be soon perfected no doubt will bring more patients to operation at a time when the malignancy is still confined to the stomach. Under such conditions, one can expect at least a 50 per cent 5-year survival rate and not infrequently a cure.

Until a better method of treating cancer of the stomach is discovered to supplant surgery, the most fertile field for improving our end results lies in the removal of all ulcerating lesions of the stomach regardless of how innocent they appear to be.

I am indebted to Dr. I. L. Tilden for his assistance with the illustrations.

REFERENCES

1. Banks, B. M., and Zetzel, L.: Prognosis in gastric ulcer treated conservatively, New England J. Med. **248:** 1008, 1953.
2. Beale, J. M., and Hill, M. R., Jr.: An evaluation of the surgical treatment of carcinoma of the stomach, Surg. Gynec. & Obst. **102:** 271, 1956.
3. Berkson, J., Walters, W., Gray, H. K., and Priestly, J. T.: Mortality of survival in cancer of the stomach: a statistical summary of the experience at the Mayo Clinic, Proc. Staff Meet. Mayo Clin. **27:** 137, 1952.
4. Blalock, J., and Ochsner, A.: Carcinoma of the stomach: a study of 18 five year survivors, Ann. Surg. **145:** 726, 1957.
5. Cain, J. C., Jordan, G. L., Jr., Comfort, M. W., and Gray, H. K.: Medically treated small gastric ulcer: five year follow-up study of 414 patients, J. A. M. A. **150:** 781, 1952.
6. Finster, H.: Ulcer—cancer of stomach, Arch. klin. Chir. **131:** 71, 1924.
7. Giberson, R. G., Dockerty, M. B., and Gray, H. K.: Leiomyosarcoma of the stomach: clinicopathologic study of 40 cases, Surg. Gynec. & Obst. **98:** 186, 1954.
8. Harvey, H. D., Titherington, J. B., Stout, A. P., and St. John, F. B.: Gastric carcinoma: experience from 1916 to 1949 and present concepts, Cancer **4:** 717, 1951.
9. Hayes, M. A.: The gastric ulcer problem, Gastroenterology **29:** 609, 1955.
10. Hitchcock, C. R., McLean, L. N., and Sullivan, W. A.: The secretory and clinical aspects of achlorhydria and gastric atrophy as precursors of gastric cancer, J. Nat. Cancer Inst. **18:** 795, 1957.
11. Kaplan, H. S., and Rigler, L. G.: Pernicious anemia and carcinoma of the stomach: Autopsy studies concerning their interrelationships, Am. J. M. Sc. **209:** 339, 1945.
12. MacDonald, I., and Kotin, P.: Biological predeterminism in gastric carcinoma as the limiting factor of curability, Surg. Gynec. & Obst. **98:** 148, 1954.
13. Malmros, H., and Hiertonn, T.: A post-investigation of 687 medically treated cases of peptic ulcer, Acta. med. scandinav. **122:** 229, 1949.
14. Marshall, S. F.: Carcinoma of the stomach, Postgrad. Med. **27:** 683, 1960.
15. Marshall, S. F.: The relation of gastric ulcer to carcinoma of the stomach, Ann. Surg. **137:** 891, 1953.
16. Martin, L., and Lewis, N.: Peptic ulcer cases reviewed after 10 years: effect of medical treatment and indications for gastrectomy, Lancet **2:** 1115, 1949.
17. McClanahan, B. J., Mitchel, J. A., and Milliken, H. E.: Carcinoma of the stomach in Hawaii, Hawaii M. J. **8:** 32, 1948.
18. McManus, R. G., and Sommers, S. C.: Significance of gastric polyps accompanying cancer, Am. J. Clin. Path. **23:** 746, 1953.
19. Mosley, M.: Leiomyosarcoma of the stomach: review of 10 cases, Am. J. Digest. Dis. **4:** 792, 1959.
20. Natvig, P., Romcke, O., and Svaar-Seljesaetero: Medical treatment of gastric and duodenal ulcer, Acta med. scandinav **113:** 444, 1943.
21. Pack, G. T., and Livingston, E. M.: Treatment of cancer and allied diseases, New York, 1940, Paul B. Hoeber, Inc., Vol. 11.
22. Ransom, H. K.: Cancer of the stomach, Surg. Gynec. & Obst. **96:** 275, 1953.
23. Strode, J. E.: Gastric ulcer: a surgical problem, A. M. A. Arch. Surg. **75:** 202, 1957.
24. Strode, J. E.: Giant hypertrophy of gastric mucosa, SURGERY **14:** 256, 1957.
25. Strode, J. E.: In support of surgical removal of small ulcerating lesions of the stomach

without benefit of medical treatment, Surg. Gynec. & Obst. **98:** 607, 1954.

26. Strode, J. E.: Observations on carcinoma of the stomach in Hawaii: a study of 140 consecutive cases coming to surgery, Hawaii M. J. **10:** 173, 1951.
27. Swynnerton, B. F., and Tanner, N. C.: Chronic gastric ulcer: comparison between a gastroscopically controlled series treated medically and a series treated by surgery, Brit. M. J. **2:** 841, 1953.
28. Swynnerton, B. F., and Truelove, S. C.: Carcinoma of the stomach, Brit. M. J. **1:** 287, 1952.
29. Tamura, P. Y., and Curtiss, C.: Carcinoma of the stomach in the young adult, Cancer **13:** 379, 1960.
30. Wangensteen, O. H.: Cancer of the esophagus and the stomach; a monograph for the physician, Am. Cancer Soc., Inc., ed. 2, pp. 128-129, 1956.
31. Welch, C., and Wilkins, E. W., Jr.: Carcinoma of the stomach, Ann. Surg. **148:** 666, 1958.
32. Wilson, L. B., and MacCarty, W. C.: Pathological relationships of gastric ulcer and gastric carcinoma, Am. J. M. Sc. **138:** 846, 1909.

Partial gastrectomy for chronic duodenal ulcer with hemorrhage: results in 450 cases*

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Hemorrhage continues to be the major and most dreaded of all complications associated with chronic duodenal ulcer. In recent years the early institution of adequate surgical intervention has been more commonly applied than before. This policy has been one of the most important steps forward in the reduction of the over-all mortality and morbidity rates. A more positive approach to this problem by all members of the medical profession should provide many more salutary results.

The following criteria were applied in practically all cases in this series for the classification of the degree of bleeding or hemorrhage:

Massive
a. 5 or more transfusions of 500 ml. of whole blood required in a period of 24 to 48 hours to restore and stabilize the circulatory blood volume, if it could be restored, or
b. hemoglobin concentration of 7.0 Gm. or less on admission.

Moderate
a. less than 5 transfusions required to restore and stabilize the blood volume in a period of 24 to 48 hours, or
b. hemoglobin concentration of 7.0 to 10.0 Gm. on admission.

Minimal
all others.

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*These results will be included in a cooperative peptic ulcer study that the Veterans Administration Research in Surgery is conducting.

PURPOSE

The purpose of this report is to present our results with this procedure and our method of management in 450 unselected and consecutive patients treated by partial gastrectomy during a 12-year period from 1946 to 1958 on our surgical service.

Basically, many factors concerned with the indications for operation, management, surgical technique employed, and follow-up studies were controlled through the direct guidance and supervision of one Chief of the Surgical Service (L. T. P.) and his staff who were trained by him. This single control standardized as far as possible most of the items which affect the over-all interpretation and results. Many of the other reports on this subject are compilations of data from several surgeons in the same institution who have conflicting and different basic tenets concerning the management of the surgical problem.

This report will point up the serious hazards associated with hemorrhage, single or repeated, in patients in all age groups. Conservative management by any group of the medical profession, as applied particularly to patients under 50 years of age, is a fallacy. Many of the dogmatic tenets which previously were universally taught and applied should be discarded.

AGE DISTRIBUTION

The average age in our series was 54.2 years. The youngest patient was 24 years

Table I. *Age distribution*

*Age**	*No.*	*%*
20-29	51	11.3
30-39	99	22.0
40-49	76	16.9
50-59	150	33.3
60-69	72	16.0

*A few patients were over 70 years of age.

Table II. *Previous surgical treatment*

Type	*No.*	*%*
Gastroenterostomy	15	3.3
Gastrectomy, partial	10	2.2
Gastrectomy, subtotal	1	0.2
Reresection	2	0.4
Perforation, closure	52	11.5
Totals	80	17.6

old and the oldest was 81. The greatest number, 33 per cent, were in the 50 to 59 year age group. However, 16.5 per cent were over 60 years old (Table I).

The number of patients under the age of 50 was equal to the number 50 or over.

DURATION OF SYMPTOMS AND PREOPERATIVE COMPLICATIONS

The average duration of symptoms prior to resection was 11.7 years. In the group of patients under 50 years of age, the duration of symptoms was 15 years, while it was 8.5 years in the group 50 or over. Eighty patients (17.6 per cent) had had previous surgical treatment (Table II).

PREOPERATIVE GASTROINTESTINAL BLEEDING

In this survey are included all of the cases in which bleeding occurred. This represents 450 patients (64 per cent) with bleeding out of a total of 700 patients who had resection for chronic duodenal ulcer during this 12-year period. It is apparent that hemorrhage or bleeding was not only the most serious complication of chronic duodenal ulcer, but also the most common.

Of the 450 patients with hemorrhage, 163 (36 per cent) were admitted to this hospital during or immediately after their initial hemorrhage. The remaining number, 287 (64 per cent), had had more than one episode of bleeding when admitted to our hospital for medical and/or surgical care.

In the entire series, 226 patients (50 per cent) were less than 50 years of age, and 224 (50 per cent) were older than 50. In the group of patients under 50 years of age, 16 per cent were treated for massive hemorrhage, as compared to 23 per cent in the group 50 or older. However, the incidence of massive hemorrhage was greatest in the group 50 to 59 years of age and is the most frequent type in the entire series (Table III and Fig. 1).

Of the 450 patients, 287 (63.7 per cent) were admitted with a history of 2 or more episodes of bleeding (Table IV). The incidence of recurring hemorrhage in the patients below 50 and those older than 50 years of age is about equal. This indicates that the chance of recurrent hemorrhage in the young patient is as great as it is in the older patient. However, the chance of massive hemorrhage is 30 per cent greater for the patient 50 or older than it is in the younger patient.

In Fig. 2 the incidence of recurrent hemorrhage is shown for each age group. This shows that recurrent hemorrhage, beyond the second episode, is more frequent in patients below the age of 50 than in patients over 50. The reversals in percentages as seen in Fig. 2 point up the fact that the young patient is and has been temporized with and not referred for operation early. Instead, he has been treated conservatively and allowed

Table III. *Age incidence and degree of bleeding*

	Massive		*Moderate*		*Minimal*	
Age	*No.*	*%*	*No.*	*%*	*No.*	*%*
20-29	17	3.9	5	1.1	29	6.4
30-39	26	5.8	26	5.8	47	10.4
40-49	27	6.0	28	6.2	21	4.7
50-59	69	15.3	30	6.9	51	11.3
60-69	34	7.5	24	5.4	14	3.1
70 or over	2	0.4	0		0	
Totals	175	39	113	25	162	36

Table IV. *Recurrent hemorrhages by age groups**

Age	Initial		Two		Three or more	
	No.	%	No.	%	No.	%
20-29	10	2.2	10	2.2	19	4.2
30-39	26	5.8	10	2.2	45	10
40-49	17	3.9	19	4.2	46	10.2
50-59	72	16	32	7.1	52	11.5
60-69	37	8.2	21	4.7	20	4.4
70 or over	1	0.2	0		13	3
Totals	163	36	92	20	195	44

*At time of surgical intervention.

to bleed again and again at later dates. The reverse appears to be true in the patient over 50 years of age. In this group the patient is referred more frequently for operation during his first episode of bleeding.

Partial gastrectomy was performed as an emergency procedure in 81 patients (18 per cent). In 101 (22.4 per cent) the procedure was performed as an immediate elective operation within 7 to 14 days after the patient was admitted to the hospital.

TYPE OF OPERATION PERFORMED

A partial gastrectomy was performed through a supraumbilical, transverse, transrectus, elliptical, abdominal incision which was closed by suturing the anterior and posterior rectus sheaths with interrupted No. 2-0 silk sutures. Approximately 75 per cent of the stomach was resected, including practically all of the lesser curvature. In all cases the duodenal ulcer was removed. An anterior gastrojejunal anastomosis was performed. In patients in whom the stomach was large and the gastric ostium wide, the lesser curvature portion of the stomach ostium was closed so that a Hofmeister type of anastomosis could be performed. The afferent loop was short in over 50 per cent of the cases and averaged 4 to 5 inches in length. The majority of the patients were ambulated on the first postoperative day.

RESULTS

The follow-up studies were conducted at 6-month intervals for the first few years, then at yearly intervals for the succeeding 2 years, and every 3 to 4 years thereafter. For the yearly and later follow-up studies, the patients were admitted to the hospital for a complete clinical and laboratory evaluation. At present follow-up is being carried out in 95 per cent of the patients. The longest follow-up period is 13 years and the shortest 22 months, with 76 per cent of group followed for over 3 years and 61 per cent followed for 5 over years. However, this study is mainly concerned with the immediate postoperative results.

POSTOPERATIVE COMPLICATIONS

There were 320 postoperative complications occurring in 299 patients (66.4 per cent). This is a much higher incidence than

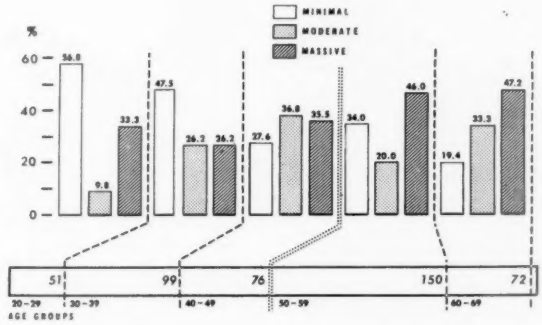

Fig. 1. Incidence of bleeding by degrees as it occurred in patients of each decade. Over 45 per cent of the patients who were over 50 when they bled hemorrhaged massively. The unlabeled portion of the illustration at the far right represents the few patients who were over 70 years of age.

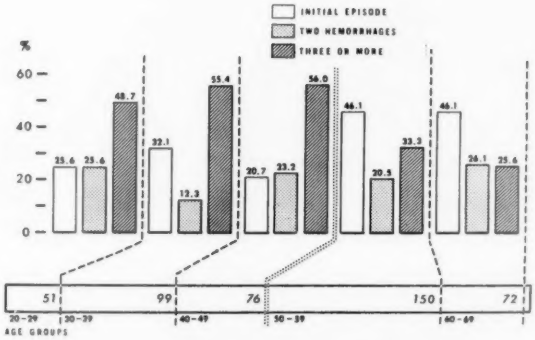

Fig. 2. Recurrent hemorrhage.

we have experienced in a large group of patients with chronic duodenal ulcer who were operated upon for reasons other than hemorrhage. The most frequent types of postoperative complications were pulmonary and vascular (Table V).

POSTOPERATIVE DEATHS

There were 25 deaths, an over-all mortality rate of 5.6 per cent in 450 patients surgically treated for hemorrhage or bleeding. The mortality rate in 175 patients operated upon because of massive hemorrhage was 9.1 per cent, or 16 deaths (Table VI and Fig. 3).

The incidence of myocardial infarction in patients under 50 years of age was unusually high in this group. In spite of this, the mortality rate in patients below 50 years of age was 50 per cent lower than in patients over 50. There were 12 deaths (11.4 per cent) in the group of 105 patients who were 50 or older as compared to 4 deaths (5.7 per cent) in the group of 70 patients who were under 50.

Hemorrhage had occurred previously in 25 per cent of the patients over 50 who died, whereas all 4 of the patients under 50 who died had had repeated hemorrhages over a period of years prior to the time they were referred for surgical consultation.

EMERGENCY OPERATIONS

Eighty-one patients (18 per cent) were operated upon as an emergency. There were 10 deaths (12.8 per cent) (Table VII and Fig. 4).

Seventy-five patients were over 50 years of age. Of this number, 8 died (10.6 per cent). There were only 6 patients under 50 years of age with 2 deaths, 1 of which was due to acute myocardial infarction. Even though the incidence of massive hemorrhage was higher in the group of patients over 50 (60 per cent), a much higher percentage (51 per cent) were operated upon as an emergency as compared to only 36 per cent in the younger age group who made up 40 per cent of the total number with massive hemorrhage (Fig. 4).

Table V. *Postoperative complications in 299 patients*

Type	*No.*	
Wound		
Dehiscence	2	
Hematoma	13	
Hernia, postoperative	7	
Infection	7	
Total	29	(6.4%)
Pulmonary		
Atelectasis	31	
Embolism	1	
Infarction	10	
Pneumonia	11	
Pneumonitis	10	
Total	63	(14%)
Vascular		
Phlebothrombosis	4	
Thrombophlebitis	19	
Postoperative hemorrhage		
Gastrointestinal tract	24	
Other	14	
Total	61	(13.5%)
Miscellaneous		
Abscess, subhepatic	6	
Cerebral hemorrhage	3	
Cholecystitis, acute	4	
Diarrhea, nonspecific	20	
Diarrhea, enterocolitis, specific	5	
Diverticulitis, acute	9	
Ileus, paralytic	4	
Jaundice, homologous serum	24	
Jaundice, obstructive	7	
Jaundice, transient	5	
Leak, duodenal stump	20	
Myocardial infarction	5	
Obstruction, efferent loop, transient	19	
Oliguria or anuria	4	
Pancreatitis, acute	3	
Pancreatitis, necrotizing	4	
Retention, gastric	15	
Uremia	5	
Urinary tract infection	5	
Total	167	(37.1%)
Grand total	320	(71.2%)

EARLY ELECTIVE OPERATIONS (7 TO 14 DAYS)

There were 101 patients (22.4 per cent) operated upon electively at an early date because of hemorrhage (Fig. 5). Operations were performed between the seventh and the fourteenth day after hemorrhage. The

mortality rate in this group was less than half of that in the emergency group.

The delay was based upon many factors which, in the judgment of the medical and surgical team, deemed it safe to elect a later time for surgical intervention.

Four deaths (9.7 per cent) occurred in the 41 patients over 50 years of age in this group as compared to 2 deaths (3.3 per cent) in 60 patients under 50. It is most unusual for a myocardial infarction to occur and to be fatal in young patients (26 and 42 years of age as noted in Table VIII). If patients with this complication were excluded from the group below 50 years of age, there would be no deaths directly attributed to the operation.

Since a higher percentage of patients over 50 years of age with massive hemorrhage were operated upon as an emergency, a smaller percentage (49 per cent) were left to be included in the group with early elective operation. However, a much larger percentage (67 per cent) of the patients in the group under 50 years of age were in the group with early elective operation—almost 3 times more than were in the group with emergency operation.

Table VI. *Mortality rate in 175 patients with massive hemorrhage*

No.	*Cause*	*Age of patient*
1	Anastomotic breakdown	58
2	Gangrene of duodenum	57, 61
4	Hemorrhage, gastric, postoperative	39, 55, 58, 61
1	Hemorrhage, hepatic artery	53
5	Myocardial infarction	26, 49, 51, 60, 60
3	Perforation, duodenal stump	31, 56, 67
Total 16	(9.1%) Average age	52.6

Table VII. *Mortality rate in 81 emergency cases*

No.	*Cause*	*Age of patient*
1	Anastomotic breakdown	58
2	Gangrene of duodenum	57, 61
2	Hemorrhage, gastric, postoperative	39, 61
1	Hemorrhage, hepatic artery	53
3	Myocardial infarction	48, 60, 60
1	Perforation, duodenal stump	56
Total 10	(12.8%) Average age	53

LATE ELECTIVE OPERATIONS (AFTER 2 WEEKS)

The group with late elective operation consisted of the remaining 268 patients or 60 per cent (Table IX and Fig. 6). There were 9 deaths, a 3.3 per cent mortality rate. This represents the lowest mortality rate of all groups reported. Pulmonary infarction occurred twice in this series of 268 patients, in both instances in patients under 50 years of age. This factor alone raises the mortality rate in the younger patient above that of the patients over 50. Otherwise the mortality rates would have been equal.

Two deaths (2.2 per cent) occurred in the 91 patients over 50 years of age in this group as compared to 7 deaths (3.9 per cent) in 177 patients under 50.

Fig. 7 summarizes the total number of patients operated upon with emergency and early elective procedures by groups, above or below the age of 50.

Fig. 7 shows a more than 50 per cent decrease in the total number of patients over 50 who were operated upon as an early elective procedure. This apparently indicates that a greater hazard is considered to exist for patients who are 50 or older and therefore these patients are operated upon earlier than patients under 50.

The late over-all results in the group of patients who survived and in whom adequate follow-ups were obtained (86 per cent) were considered excellent to good in 83 per cent.

COMMENT

Because this group of 450 patients was carefully controlled from every aspect over

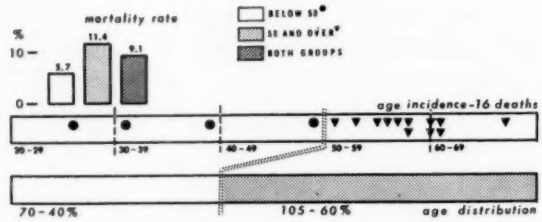

Fig. 3. One hundred seventy-five cases of massive hemorrhage.

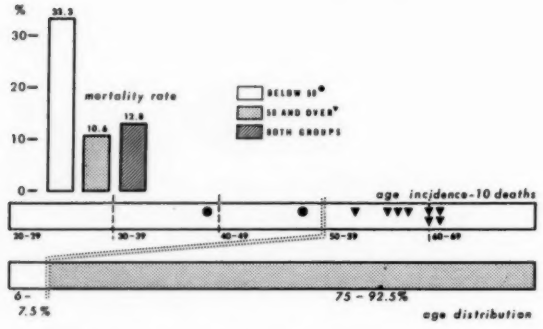

Fig. 4. Eighty-one emergency operations.

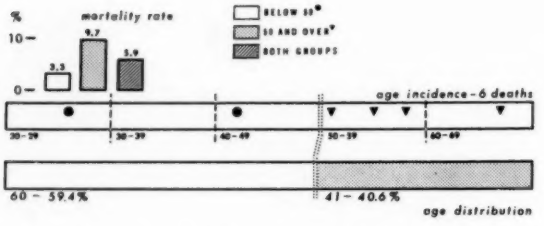

Fig. 5. One hundred one early elective operations.

a 12-year period, and because the number of patients in the age groups above and below 50 years were equal, the evaluation of the recorded data in a comparative manner provides us with many important statistics that can be used as guide lines in the future management of patients with hemorrhage from a chronic duodenal ulcer.

The incidence of bleeding or hemorrhage remains as the most frequent and serious complication of chronic duodenal ulcer. Massive hemorrhage occurs more frequently in all age groups than bleeding of lesser degrees. As patients become older the incidence and possibly the severity of bleeding increase. However, it must be stressed that the young patient with massive hemorrhage can be as vulnerable to the many hazards associated with hemorrhage as the older patient. They can become exsanguinated almost as frequently and as readily as an older patient even though the younger patient is capable, because of his youth, of withstanding more readily the insults of repeated shock.

In this respect, many physicians, even today, procrastinate more frequently with young patients who are bleeding. The risks involved in such a policy are greater than most physicians realize. Even though these patients may survive 1 or 2 episodes of bleeding, as they become older the ravages of aging and the repeated episodes of shock take their toll. When such patients are finally referred for operation the morbidity and mortality rates are much higher. This is apparent in our series.

The problem is clearly seen in our series of patients. For example, the duration of symptoms prior to the institution of definitive surgical intervention was 15 years for patients under 50 years of age as compared to 8.5 years for the older patient.

However, we have seen in the later years of our study, that more patients in the younger age groups are being referred to us earlier and during their current bleeding episodes. Therefore, surgical intervention is practiced more frequently and also earlier than before. This has resulted in a lowering of the morbidity and mortality rates and the prevention of the deterioration of the patient to a point of irreversible damage or death.

With the benefits of seasoned surgical judgment, broader surgical experience, organization of the surgical and allied teams,

Table VIII. *Mortality rate in 101 patients with early elective operation (7-14 days)*

No.	*Cause*	*Age of patient*
2	Hemorrhage, gastric, postoperative	55, 58
3	Myocardial infarction	26, 42, 51
1	Perforation, duodenal stump	67
Total 6	(5.9%) Average age	49.5

Table IX. *Mortality rate in 268 patients with late elective operation*

No.	*Cause*	*Age of patient*
5	Acute necrotizing pancreatitis	31, 32, 41, 48, 52
1	Hepatic failure	64
2	Infarct, pulmonary	28, 36
1	Perforation, duodenal stump	31
Total 9	(3.36%) Average age	40.2

the free use of whole blood, and closer cooperation between physicians and surgeons, a further decrease in both morbidity and mortality rates can be expected. In conjunction with this, better technical skill and experience will reduce the frequency of technical deficiencies and eliminate many of the intra-abdominal complications that frequently cause death.

The mortality rate was 50 per cent lower in patients under 50 years of age who were operated upon for massive hemorrhage. This is further evidence that an arbitrary age, such as 50 years or over, should not be the main guiding line in influencing the decision for surgical intervention. Instead, surgical intervention should be based upon individual judgment in each specific case and it should be remembered that the young patient is vulnerable to all the hazards associated with bleeding. Since recurrent hemorrhage can be expected in a high percentage of these patient, it is unwise to allow them to be subject to the recurrent disability associated with chronic duodenal ulcer as they become older and to finally have them reach an age when the operative risks become greater. These greater risks are due to many factors associated with aging and intercurrent diseases.

In this study, there was a marked difference in the percentages of patients operated upon as an emergency based upon age. Seventy per cent were over 50 years of age as compared to only 8.5 per cent under 50. This fully bears out the comments noted. During the first several years included in this survey, a high percentage of young patients with hemorrhage were not seen by or referred to the surgeon unless they were in dire circumstances. This accounts in part for the marked discrepancy in the percentage of patients by age groups coming to emergency operation. In recent years, with a better liaison and clearer understanding of this problem with our medical colleagues, the indications for surgical intervention are now applied along sound medical and surgical principles without serious or sole regard of the patient's age.

The mortality rate in patients with emergency operations was higher than in any of the other groups, as shown in the various mortality rate tables. This rate was lower than in former years and can be expected to show a decline in later years. In this group

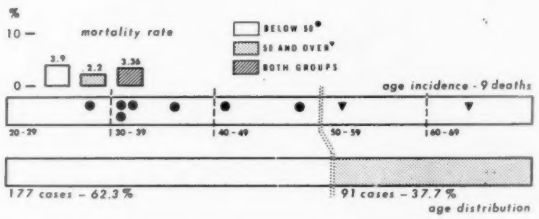

Fig. 6. Two hundred sixty-eight late elective operations.

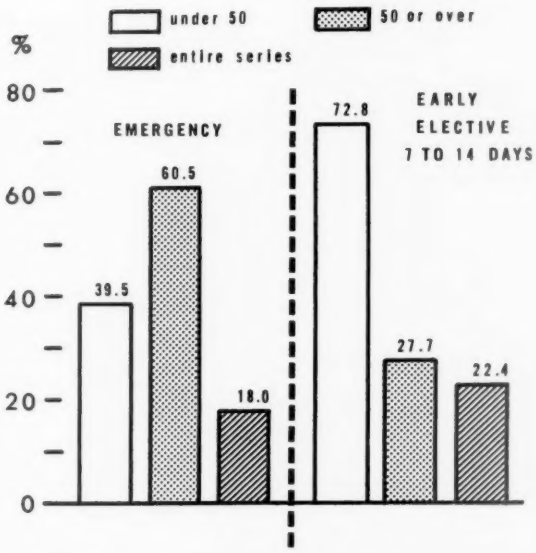

Fig. 7. Patient age versus surgical status.

were the poor-risk patients, many of whom had sustained considerable blood loss either quickly or over a long period of time. Many of them had been referred for surgical consultation late in their current emergency. All these factors pose a grave responsibility for the surgeon who is called upon to attempt to save the patient's life.

The mortality rate showed a favorable decline when operation was delayed. However, this was a group of patients in whom the medical-surgical team was reasonably sure that exsanguination and death or profound, prolonged, irreversible shock would not occur. Therefore, by waiting for a period of time, they were able to prepare the patients more adequately for elective surgical intervention, usually 7 to 14 days after hemorrhage. This policy is reflected in a mortality rate of 5.9 per cent. This is 50 per cent lower than in the group who were operated upon as an emergency. In the group who underwent late elective operation, the morbidity and mortality rates were lowest. This is expected since operation can be accomplished under most favorable circumstances, when there is no hurry or tension and fewer hazards exist. Also, a higher percentage (66 per cent) of the patients were under 50 years of age.

SUMMARY AND CONCLUSIONS

Hemorrhage or bleeding from a duodenal ulcer is the most frequent, the most serious, and the most dreaded of all complications. It results in the highest morbidity and mortality rates of all the complications associated with this lesion. The mortality rate is greatly influenced by the degree and the duration of hemorrhage, the age of the patient, the degree of liaison between physician and surgeon, the effectiveness of organized positive treatment and ancillary supportive measures, surgical judgment and experience, and the surgical procedure performed.

The mortality rate is highest in the group of patients operated upon as an emergency. In this group, however, a progressive decline in the death rate is occurring. This is due to many factors. One of the most important is earlier surgical intervention. Another is the recognition by both the medical and surgical groups that age is no longer the main determining factor for surgical intervention. Hemorrhage in patients under 50 years of age can be as serious and as lethal as that in patients over 50. Recurrent hemorrhage is frequent in all age groups and massive hemorrhage is the most frequent type.

A survey of 450 consecutive and unselected patients treated for hemorrhage from a chronic duodenal ulcer during a 12-year period (1946 to 1958) on our surgical service from a total of 700 patients operated upon for chronic duodenal ulcer substantiates our conclusions.

Persistent conservative management in the face of all the hazards associated in the long-term history of a chronic duodenal ulcer is a policy which most frequently results in prolonged disability and finally in high morbidity and mortality rates associated with hemorrhage.

Management of hemorrhage, in all age groups, requires a more positive medical and surgical approach. Through such a program, along with the application of seasoned judgment with regard to each individual patient, we can expect a more realistic final outcome. This should include a lowering of the morbidity and mortality rates to a point almost equal to those now reported for the other complications of a chronic duodenal ulcer which require surgical treatment.

Factors which effect release of pepsinogen granules from gastric tubules

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Pepsinogen granules are the precursors of the proteolytic enzyme pepsin and are stored in the chief cells of the gastric glands. In 1874, Grützner[6] theorized the existence of a precursor of pepsin in the fundic portion of the stomach. However, Langley and Sewall,[9] in 1879, were the first to observe these granules in the gastric mucosa of different animals. They observed that the granules accumulated in the gastric glands during fasting and were released on feeding. It has been shown also that pilocarpine produces marked discharge of pepsinogen.[5, 12] Other studies of gastric secretion have shown that histamine stimulates acid production, but there has been controversy concerning it effect on pepsin secretion.[2, 4, 7] Vagotomy regularly inhibits gastric secretion.[1] Hyperventilation has been shown to decrease gastric secretion while hypercapnea produces the opposite effect.[3, 8] A specific pattern of gastric secretion after cerebral trauma has been demonstrated experimentally.[10] Studies of the effect of temperature changes on gastric secretion have shown that a marked decrease occurs with hypothermia.[11]

It is the purpose of this presentation to report our observations of the histologic appearance of the gastric mucosa when the environmental changes occur. The effect of gastric hypothermia upon release of pepsinogen granules under these same conditions also has been studied.

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METHODS AND MATERIALS

Staining technique. The first problem encountered was in devising a staining method capable of demonstrating pepsinogen granules in the chief cells of the gastric glands. A modification of the thionine stain described by Lillie in 1954 was used. The gastric biopsy specimens were placed in Kose's fixative (9 parts 3 per cent potassium bichromate, 1 part 37 per cent formalin) and sun-yellow dye was used as a counterstain. The granules appeared blue to purple in the stomachs of the various animals examined.

ANIMAL EXPERIMENTS

The animals used in this experiment were dogs, cats, and rabbits. In all animals, A 48-hour fasting period was enforced prior to the beginning of any experimental procedure. Because rabbits retain stomach contents despite prolonged fasting, gastric lavage was used before the standard 48-hour fasting period. In dogs, whenever anesthesia was necessary to obtain biopsy specimens, urethane (1 Gm. per kilogram of body weight) was administered intravenously. In cats and rabbits, Nembutal (30 mg. per kilogram) was used. The amount of pepsinogen granulation observed after a 48-hour fast was

Table I. *Effect of various stimuli and inhibitors on the concentration of the pepsinogen granules in the chief cells of the stomach in dogs, cats, and rabbits*

Type of experiment	*No. of dogs*	*No. of cats*	*No. of rabbits*	*Change in concentration of granules in cells*
1 48 hr. fasting	3	3	3	Moderate concentration of granules in cells
2 Pilocarpine	3	3	3	Marked decrease in concentration of granules
3 Histamine (0.5 mg.)	3	3	3	Moderate increase or no change in concentration of granules from that in fasting specimens
4 Histamine (2.5 mg.)	3	3	3	Moderate decrease in concentration of granules
5 Histamine (10 mg.)	3	—	—	Moderate decrease in concentration of granules
6 Atropine	3	3	3	Moderate increase or no change in concentration of granules from that of control specimen
7 After feeding	3	3	—	Marked decrease in concentration of granules
8 Local gastric cooling	3	3	3	Marked increase in concentration of granules
9 Aftereffect of cooling	3	3	—	Marked decrease in concentration of granules
10 Pilocarpine and local gastric cooling	3	3	—	Marked increase in concentration of granules in cell, similar to local cooling
11 Local gastric hyperthermia	3	3	3	Marked increase in granule concentration
12 Hyperventilation	3	—	—	Marked increase in concentration of granules
13 Head trauma	3	—	—	
Before compression				Similar to 48 hr. fasting specimens
4 hr. after compression				Marked increase in granule concentration
24 hr. after compression				Marked decrease in concentration of the granules in cells
14 Vagotomy (2 wk.)	3	—	—	Moderate to marked increase in concentration of granules in cells
15 Vagotomy prior to feeding	3	—	—	Moderate decrease in concentration of granules in cells
16 Vagotomy (3 mo.-24 mo.)	2	—	—	Moderate to marked increase in concentration of granules in cells
17 Vagotomy prior to feeding	2	—	—	Moderate decrease in concentration of granules in cells

established for each animal. Any decrease or increase in the number of pepsinogen granules noted after the institution of the experimental conditions was interpreted as the specific effect of the agent employed.

Stimulation of release of pepsinogen granules by feeding was noted. The observations were made on cats which received 150 Gm. of canned cat food, and on dogs which received 250 Gm. of beef. Exactly 4 hours after feeding, the animals were sacrificed and biopsies of specimens of the fundic portions of the stomach were obtained immediately. Fasting dogs and cats were given pilocarpine (3.2 mg.) subcutaneously every hour for 4 hours and gastric biopsies were taken 1 hour after the last dose had been given. Similar stimulation was carried out with histamine, 0.5 and 2.5 mg., given every hour for 4 hours. One hour after the last dose of histamine had been given, a gastric biopsy was taken. In addition, a single 10 mg. dose of histamine, after Benadryl protection, was administered and biopsies were taken at the end of 4 hours. Gastric specimens were taken after the subcutaneous ad-

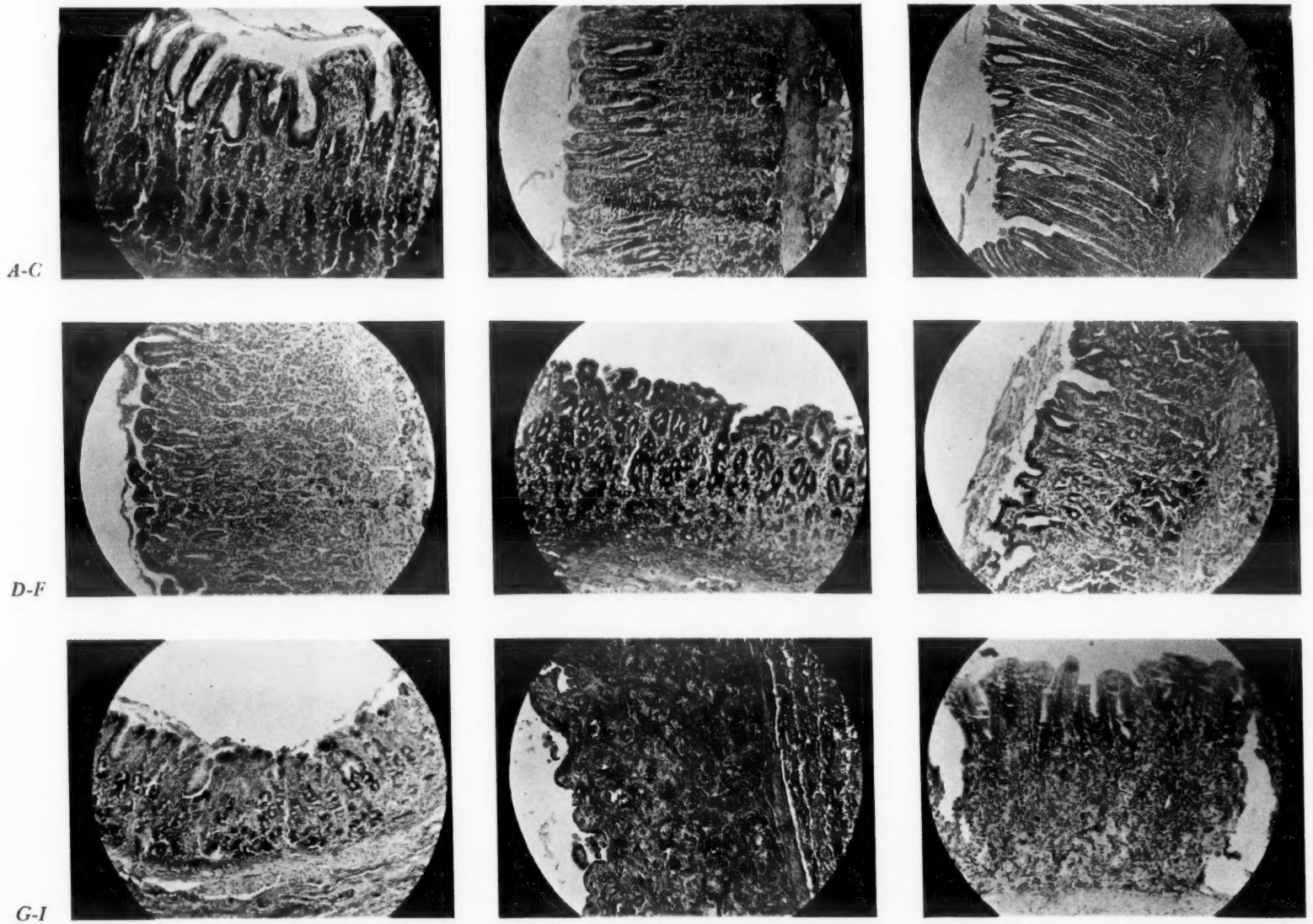

Fig. 1. *A*, Gastric mucosa from dog fasted for 48 hours. *B*, Four hours after feeding. *C*, After 4 injections of pilocarpine, 3.2 mg. *D*, After 4 injections of histamine, 2.5 mg. *E*, During hypothermia. *F*, Cold stomach stimulated 3 times with 3.2 mg. pilocarpine. *G*, Three hours after cooling. *H*, During hyperthermia. *I*, Dog fasted for 48 hours. Fifteen days after vagotomy. (Original magnification ×135 throughout.)

ministration of 0.13 mg. of atropine every hour for 4 hours.

After a preliminary biopsy specimen had been taken, local gastric hypothermia was provoked in dogs, cats, and rabbits by the circulation of fluid at –5° C. through an inlying gastric intraluminal balloon for 3 hours. Biopsy specimens were then obtained from the stomach and the cooling was stopped. Three hours later, with the stomach at 38.5° C., a third specimen was taken for comparison. In one series, pilocarpine was administered during the cooling and a biopsy specimen was obtained 1 hour later, when the stomach wall was still cold.

In addition, the effects of hyperthermia, vagotomy with and without feeding, hyperventilation, hypercapnea, and cerebral compression produced by accelerating head trauma were examined in various animals.

RESULTS

The pepsinogen granules, stained by the method we employed, varied in color from animal to animal. They were stained bluish purple in the dog, purple in the cat, and blue in the rabbit. An unstimulated chief cell showed granulation of varying degrees with a small eccentric nucleus. Frequently the cell boundaries were fused with those of neighboring cells. The secreting cell had no granules in the cytoplasm, which was often vacuolated. The cell was shrunken and the nucleus appeared relatively large.

The 48-hour fasting specimen, considered a resting mucosa, showed moderate concentration of pepsinogen granules in the cytoplasm of the chief cells, which were not maximally distended. The cells with granules were located predominantly in the deeper portions of the gastric glands, and the nuclei were darkly stained and clearly seen. Fig. 1, *A* shows the typical histologic appearance of gastric mucosa in the dog after 48 hours of fasting.

The results of the various experimental procedures are shown in Table I.

Stimulation. Feeding stimulated maximal discharge of the pepsinogen granules. The chief cells 4 hours after feeding were seen to be shrunken, cuboidal in shape, and with a clear cytoplasm. The nucleus was relatively larger (Fig. 1, *B*). Similarly, pilocarpine produced a marked decrease in the number of pepsinogen granules and most of the chief cells were devoid of granules and had a small vacuolated cytoplasm and large nuclei (Fig. 1, *C*). This correlated well with the secretion of gastric juice of high enzyme content. Small doses of histamine (0.5 mg. per hour) produced no significant change in the granule concentration from that present during the fasting state. On the other hand, all animals which received large doses of histamine (2.5 mg. per hour, 10 mg. dose) showed a decreased concentration of gran-

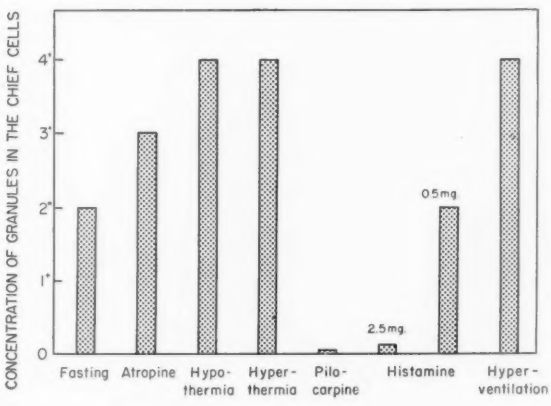

Fig. 2. Concentration of pepsinogen granules in the chief cells after 4 hours of stimulation with various stimulants.

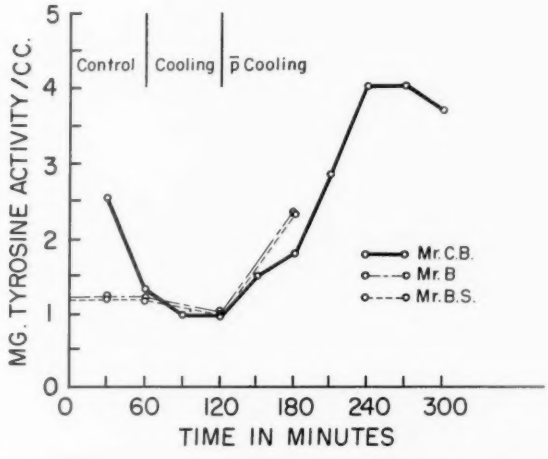

Fig. 3. Pepsin concentration in patient before, during, and after cooling.

Table II. *Changes in pepsin and gastric juice volume in dogs subjected to cerebral compression*

Dog No.	*Pepsin* (mg./ml.)*	*pH*	*F. A.*	*Volume*	*Change of volume† (ml./half hour)*
1 934	→	↓	↑	↑	0.4 - 14
2 767	↑ 0.4 - 0.8	↓	↑	↑	0.3 - 2
3 677	↑	↓	↑	↑	0.4 - 7
4 927	↑ 0.4 - 0.6	↓	↑	↑	1.8 - 16
5 1282	→	↓	↑	↑	3.0 - 9.5
6 1156	→	↓	↑	↑	4.0 - 10.6
7 1346	→	↓	↑	↑	3.0 - 4
8 1272	↑ 0.2 - 0.3	↓	↑	↑	8.0 - 14
9 1509	↑ 0.45 - 0.6	→	↑	↑	4.0 - 10.2
10 1586	↑ 0.2 - 0.6	↓	↑	↑	0.7 - 7.25
11 1442	→	↓	↑	↑	0.5 - 6
12 1897	→	↓	↑	↑	4.0 - 4.7
13 199	↑ 0.2 - 0.3	↓	↑	↑	2.4 - 27.5
14 226	↑ 0.4 - 0.65	→	→	↑	0.7 - 4
					33.2 136.7
					Mean 2.4 - 9.7

*Pepsin was measured by milligrams of tyrosine activity per milliliter gastric juice.
†Average increase in volume of gastric juice after trauma = 7.3 ml. in ½ hr.
↓ Decrease in amount.
↑ Increase in amount.
→ No change.

ules similar to that present after pilocarpine administration (Fig. 1, *D*, and Fig 2).

Inhibition. Atropine given to animals fasted for 48 hours caused an accumulation of the pepsinogen granules within the chief cells. The cells were swollen and the granules were large and tightly packed, which resulted in the nucleus being pushed into an eccentric position (Fig. 2). In addition, most of the cells showed granulation. This correlated with the decreased secretion of pepsin from pouches noted when dogs were given atropine.

Hypothermia produced a marked suppression of release of pepsinogen granules from the chief cells. After 4 hours of local cooling at 10 to 15° C., most of the chief cells were in a state of inactivity, distended to the maximum limit with pepsinogen granules, and the nucleus was small in size. The cells had a wide margin of cytoplasm which was filled with the granules and were so distended that their boundaries were indistinct (Fig. 1, *E*, and Fig. 2). In Fig. 3 one can see that this histologic picture corresponds to the secretion of pepsin.

When cooling was stopped and the gastric wall temperature had been normal (38.5° C.) for 3 hours, the chief cells were functioning maximally. The granules were gone and the cytoplasm was clear and vacuolated. The

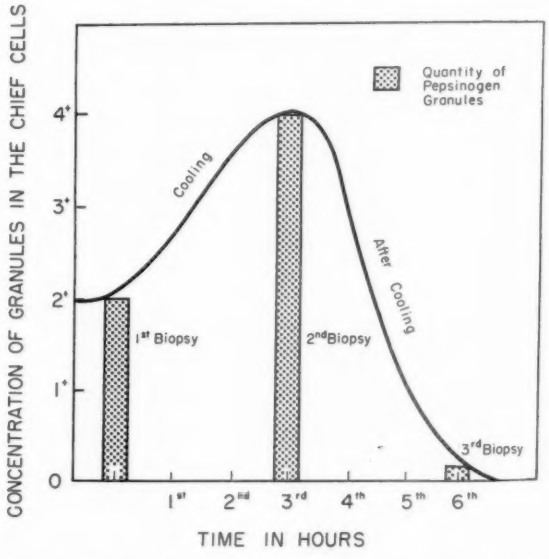

Fig. 4. Effect of local gastric cooling and aftereffect of cooling on concentration of pepsinogen granules in the chief cells.

cell was shrunken with a seemingly large nucleus (Fig. 1, *F*, and Fig. 4). Interestingly enough, while local hypothermia is in effect, pilocarpine cannot cause release or dislodgment of the granules. Specimens taken of the cold gastric mucosa (10 to 15° C.) after maximal stimulation with pilocarpine showed accumulation of pepsinogen granules with all the other signs of inactivity (Fig. 1, *G*). Hyperthermia also caused marked suppression of pensinogen granule release (Fig. 1, *H*, and Fig. 2.)

Miscellaneous observations.

Vagotomy. The effects of vagotomy on the release of pepsinogen granules from the chief cells are interesting. Two weeks after va-

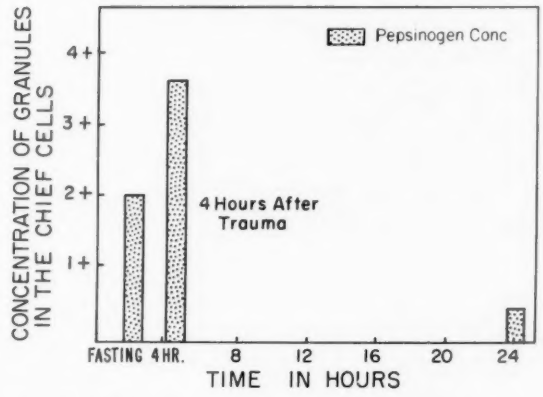

Fig. 5. Effect of cerebral compression on the concentration of pepsinogen granules in the chief cells.

Table III. *Semiquantitation of changes in concentration of pepsinogen granules in the chief cells under various conditions*

Animal	*No.*	*Procedure*	*No. of hours of stimulation*	*Concentration of granules*
Dog				
	3	48 hr. fasting		2+ (moderate)
	3	Atropine	4	3+
	3	Pilocarpine	4	0-1+
	3	Histamine, 0.5 mg. dosage	4	2+-3+
	3	Histamine, 2.5 mg. dosage	4	0-1+
	3	Histamine, 10.0 mg. dosage	1	0-1+
	3	Local cooling	4	4+
	3	Aftereffect of cooling	3	0-1+
	3	Pilocarpine and local gastric cooling	3	3+-4+
	3	Local heating	4	4+
	3	Hyperventilation	4	4+
	3	Prior cerebral compression	4	3+-4+
	3	After cerebral compression	24	0-1+
	3	Prior vagotomy	—	3+-4+
Cat				
	3	48 hr. fasting		2+ (moderate)
	3	Atropine	4	3+
	3	Pilocarpine	4	0-1+
	3	Histamine, 0.5 mg. dosage	4	2+-3+
	3	Histamine, 2.5 mg. dosage	4	0-1+
	3	Local cooling	4	4+
	3	Aftereffect of cooling	3	0-1+
	3	Pilocarpine and local gastric cooling	3	3+-4+
	3	Local heating	4	4+
Rabbit				
	3	48 hr. fasting		2+ (moderate)
	3	Atropine	4	3+
	3	Pilocarpine	4	0-1+
	3	Histamine, 0.5 mg. dosage	4	2+-3+
	3	Histamine, 2.5 mg. dosage	4	0-1+
	3	Local cooling	4	4+
	3	Local heating	4	4+

gotomy, fasting specimens showed a definite increase in the concentration of granules over that in the original biopsy specimen. Response to food was decreased but nevertheless some outpouring of granules did occur. In specimens taken from stomachs with vagal interruption of longer duration, the pattern was one of gradual partial return to normalcy. Even 2 years after vagotomy, however, fasting specimens still appeared to have a greater number of granules within the chief cells than is observed normally during the fasting state. Feeding of the vagotomized dogs caused a decrease in the granulation and some cells showed good activity, although many cells still retained the pepsinogen and did not appear stimulated at all (Fig. 1, *I*).

Hyperventilation. A marked supression of pepsinogen release in the chief cells which were distended to their full limit with granules was caused by hyperventilation, whereas hypercapnea had a moderate stimulatory effect on the chief cells (Fig. 2).

Cerebral compression. Compression produced by head trauma produced a characteristic pattern. A specimen taken 4 hours after trauma showed inactive chief cells distended with granules, with indistinct borders. However, 24 hours after the trauma the chief cells were completely devoid of pepsinogen and exhibited all the usual evidence of marked activity. This histologic appearance correlated well with the production of a gastric juice of high peptic activity (Fig. 5 and Table II).

Table I demonstrates the results of various experimental procedures in different animals. An attempt was made to grade the concentration of the granules. A 48-hour fasting specimen is characterized as a 2+ concentration of pepsinogen within the chief cells. Maximal concentration was denoted as 4+, a moderate increase over the fasting state as 3+, a moderate decrease in the fasting level as 1+, and virtual absence of granules as zero (0). The various sections studied during this investigation were all graded and recorded as shown in Table III.

SUMMARY

The effects of various changes in environmental conditions on the release of pepsinogen granules have been demonstrated. Local gastric hypothermia does not influence the formation of pepsinogen granules but inhibits their release.

REFERENCES

1. Book, T. D., Chin, N. N., Austin B., and Beams, A. J.: Studies on pepsin secretion: effect of vagal resection for duodenal ulcer, Gastroenterology **20:** 458, 1952.
2. Bowie, D. J., and Vineberg, A. M.: The selective action of histamine and the effect of prolonged vagal stimulation on the cells of the gastric glands in the dog, Quart. J. Exper. Physiol. **25:** 247, 1935.
3. Browne, J. S. L., and Vineberg, A. M.: The interdependence of gastric secretion and the CO_2 content of the blood, J. Physiol. **75:** 345, 1932.
4. Bucher, G. R., Ivy, A. C., and Gray, J. S.: Is histamine able to maintain an augmented pepsin response comparable to that of pilocarpine? Am. J. Physiol. **132:** 698, 1941.
5. Giltlitz, A. J., and Levinson, W.: The changes within the cells of the gastric mucosa during secretory activity, Am. J. Digest. Dis. **3:** 756, 1936-1937.
6. Grützner, P.: Über eine neue Methode Pepsinmenges colorimetrisch zu bestimmen, Arch. f. d. ges. Physiol. **8:** 452, 1874.
7. Hirschowitz, B. I., London, J. L., and Pollard, N. M.: Histamine stimulation of gastric pepsin secretion, Gastroenterology **32:** 85, 1957.
8. Hirschowitz, B. I., London, J. L., and Wiggins, H. S.: Stimulation of gastric pepsin secretion in man by hyperventilation, J. Lab. & Clin. Med. **53:** 577, 1959.
9. Langley, J. N., and Sewall, H.: On the changes in pepsin forming glands during secretion, J. Physiol. **2:** 281, 1879-1880.
10. Salmon, P. A., Griffen, W. O., Jr., Paterson, J. S. R., Castaneda, A., and Wangensteen, O. H.: Experimental studies in cerebral trauma and gastric secretion, S. Forum **10:** 757, 1959.
11. Wangensteen, O. H., Salmon, P. A., Griffen, W. O., Jr., Paterson, J. S. R., and Fattah, F. N.: Studies of local gastric cooling as related to peptic ulcer, Ann. Surg. **150:** 346, 1959.
12. Weber, J.: The basophilic substance of the gastric chief cells and its relation to the process to secretion, Acta Anat. **33:** 1958.

Hiatal hernia as a complication of the Heller operation

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The linear esophagomyotomy of the lower esophagus, usually referred to as the Heller operation, has become the most widely adopted procedure in the surgical treatment of achalasia. The simplicity of the operation and its effectiveness have brought about this universal acceptance. While this method of treatment is eminently successful in relieving the dysphagia caused by the resistance at the inferior esophagus, it does nothing to alter the physiologic derangement of the remainder of the organ, and, if improperly performed, it may produce effects more disturbing than dysphagia.

Esophagomyotomy has been performed at the Graduate Hospital on 48 patients with achalasia. Thirty-five of these have been followed postoperatively for from 3 to 10 years. One of us previously reported good to excellent results in 79 per cent of patients who had achalasia and in whom the Heller operation had been carried out.[6] In this study it was established radiologically that reflux of gastric contents into the esophagus occurred in each case in which the surgical result had been unsatisfactory. Others have referred to the important role played by reflux of gastric contents in the production of poor results after esophagomyotomy and have stressed the importance of careful attention to technical details at the time of operation so as to avert resultant cardioesophageal incompetence.[2, 3, 4]

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Our experience with 4 patients who had achalasia and who developed hiatal hernias after esophagomyotomy should serve to illustrate the necessity for restoration of the normal anatomic relations of the esophagus and diaphragm.

CASE REPORTS

Case 1. J. B., a 17-year-old white girl, was admitted to Graduate Hospital, July 7, 1954, with a clinical diagnosis of achalasia. The symptoms of dysphagia and daily vomiting had existed for 3 years. Hydrostatic dilatation of the esophagus had been performed at another hospital 2 years prior to admission and had been followed by only temporary relief of symptoms. After this the patient had been treated with bougienage every 2 months without lasting improvement.

On July 8, 1954, roentgen examination showed a markedly dilated and elongated esophagus with retention of 95 per cent of the barium meal at the end of 1 hour. The terminal esophagus exhibited marked conical narrowing. Peristaltic activity was not noted in the distal two-thirds of the esophagus (Fig. 1). An esophagoscopic examination on July 12, 1954, confirmed the roentgen diagnosis of achalasia.

On July 14, 1954, an esophagomyotomy was carried out through an abdominal approach. The esophagus was mobilized by division of its attachments to the diaphragm. The hiatal opening was regarded as normal and no attempt was made to narrow it.

The patient made a satisfactory recovery from the operation and within a week she tolerated a full diet without dysphagia.

Postoperative roentgen study, July 23, 1954, showed a large paraesophageal hernia (Fig. 2).

The patient remained well until Feb. 9, 1957, when she was admitted with hematemesis. She had

Fig. 1. Case 1. Preoperative roentgenogram of distal esophagus and stomach which demonstrates smooth, conical narrowing of distal esophagus with marked dilatation proximal to narrowed segment. *Dotted lines* indicate the diaphragm.

Fig. 2. Case 1. Roentgenogram made after Heller operation which shows decrease in caliber of esophagus. Note moderate hiatal hernia of stomach extending above diaphragm *(dotted lines)*.

had no dysphagia since the Heller operation but had developed severe substernal burning distress 1 month prior to this admission. The pain was most severe at night or when she was in the recumbent position and was relieved by the ingestion of milk.

On Feb. 14, 1957, roentgen examination demonstrated an increase in size of the hernia and an ulcer in the terminal esophagus (Fig. 3).

On Feb. 21, 1957, a transthoracic repair of the hiatal hernia was carried out. The hiatus was found to be markedly enlarged.

Roentgen examination on March 4, 1957, confirmed the reduction of the hernia and free passage of esophageal contents into the stomach (Fig. 4).

The patient has had no symptoms indicative of esophageal or gastric disease since.

Case 2. R. S., a 44-year-old white man, was admitted to Graduate Hospital on Feb. 14, 1955, with a diagnosis of achalasia. The onset of symptoms of dysphagia had been in 1941. Bougienage had been carried out repeatedly with no relief. The man's alcoholic intake was excessive. In recent years, daily vomiting had occurred 30 minutes after each meal. There had been a weight loss of 40 pounds.

The presence of achalasia was noted on roentgen examination on Feb. 16, 1955. This study demonstrated marked esophageal dilatation down to the level of the diaphragm. The dilated esophagus then tapered smoothly to a stenotic area through which only a trickle of barium passed. No peristaltic activity was observed in the entire esophagus (Fig. 5).

Esophagoscopic findings were consistent with the diagnosis of achalasia.

A transabdominal esophagomyotomy was carried out on Feb. 18, 1955. The esophagus was mobilized by detachment from the diaphragm. An esophagomyotomy and a complementary pyloromyotomy were performed. The hiatus was regarded as normal and no attempt was made to narrow it or fix the esophagus to it.

The patient's early postoperative course was satisfactory; dysphagia disappeared and he began to gain weight.

Four months later he noted a different type of distress which occurred in the form of severe, burning epigastric pain which would awaken him from sleep.

Roentgen examination showed a sliding hiatal hernia associated with esophagitis and ulceration (Fig. 6). This was confirmed by esophagoscopic examination.

To date, this problem has been managed by medical measures with limited success. The patient has had 5 subsequent hospital admissions, 2 of which were required for gastrointestinal bleeding. Surgical repair of the hernia has not been accepted

by the patient even though the symptoms of esophagitis are more distressing than the dysphagia that existed prior to the esophagomyotomy.

Case 3. W. R., a 38-year-old white man, was admitted to Graduate Hospital on Aug. 30, 1955, with a clinical diagnosis of achalasia. He had had dysphagia for 5 years for which he had been given anticholinergic and sedative drugs.

A dilated esophagus with a narrowed terminal segment was noted at roentgenographic examination made on Aug. 30, 1955 (Fig. 7).

On Sept. 2, 1955, an esophagomyotomy and a pyloromyotomy were carried out via an abdominal approach. The esophagus was mobilized by division of the peritoneum at its reflection from the hiatal edges. Nothing abnormal was noted at the hiatus and no effort was made to narrow it.

The early postoperative course was without incident and the patient was able to take a full diet without dysphagia.

Postoperative roentgen examination performed on Oct. 25, 1955, demonstrated a large hiatal hernia. Marked regurgitation of gastric contents could be produced by placing the patient in the Trendelenburg position (Fig. 8).

He has remained free of dysphagia but episodic burning pain is noted in the epigastrium.

Case 4. M. S., a 65-year-old white woman, was admitted to Graduate Hospital April 7, 1958, with achalasia. Dysphagia had first been noted in 1944. Dilatation of the esophagus had been performed on a number of occasions and on one of these, in 1952, the cervical esophagus had been perforated. Symptoms persisted and the woman was hospitalized for surgical treatment of achalasia. Roentgen examination showed a dilated esophagus with a conical narrowing at the cardioesophageal junction which never distended (Fig. 9).

On April 9, 1958, a Heller operation and a pyloroplasty were carried out. The peritoneal reflection about the hiatus was divided and the terminal thoracic esophagus was pulled down into the abdomen. The hiatal opening was regarded as normal and no reconstruction was performed. The postoperative course was uneventful and the patient was asymptomatic until March, 1959. At this time she had a recurrence of dysphagia, regurgitation, and vomiting.

She was re-admitted on Aug. 13, 1959. At this time, weight loss was evident. Roentgen examination on Aug. 15, 1959, demonstrated esophageal retention and dilatation and a small hiatal hernia (Fig. 10). An esophagoscopic examination confirmed the marked narrowing of the terminal esophagus and suggested the presence of an ulcer in this segment. These findings were substantiated by another roentgen study (Fig. 11).

On Sept. 2, 1959, a transthoracic exploration of the lower esophagus and stomach was performed. An irreducible hiatal hernia was demonstrated and, at the gastroesophageal junction, there was a large ulcer. The distal esophagus was resected and continuity was restored by interposition of a segment of jejunum. Bilateral vagotomy was also carried out. Postoperative roentgen exam-

Fig. 3. Case 1. Marked increase in size of hernia demonstrated 2½ years later.

Fig. 4. Case 1. Postoperative roentgenogram made after reduction and repair of hiatal hernia.

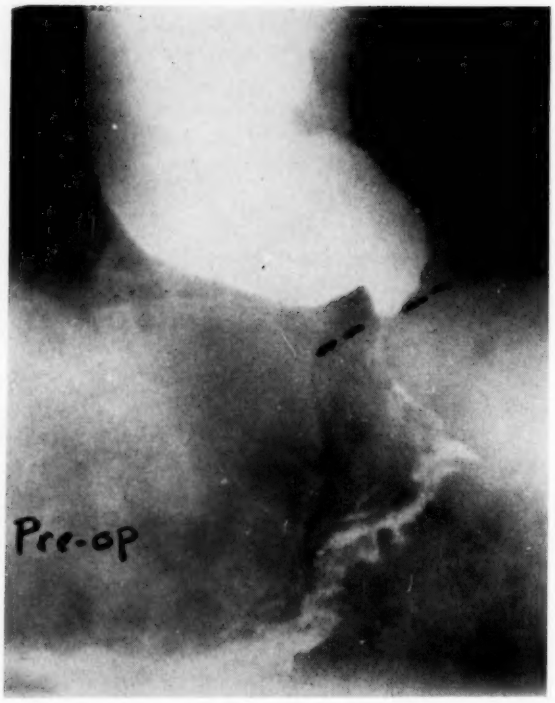


Fig. 5. Case 2. Preoperative roentgenogram which demonstrates marked esophageal dilatation down to level of diaphragm *(dotted lines)* where marked conical constriction begins.

Fig. 6. Case 2. Film study which shows hiatal hernia made after Heller procedure.

ination showed rapid emptying of the esophagus through the interposed segment (Fig. 12). Convalescence has been most satisfactory and the patient is gaining weight.

COMMENT

In the cases just described, either the achalasia and hiatal hernia were coexistent prior to the operation and were not recognized by the roentgenologist, esophagoscopist, and surgeon in each case, or the hiatal hernia was the result of the operative manipulation. It is our belief that the latter is the more likely possibility.

Until recently very few references were made to the simultaneous occurrence of achalasia and hiatal hernia. In 1957 Euphrat and Delmonico[5] recorded a case which they regarded as the third reported example of achalasia coexisting with hiatal hernia and they quoted several other authors who also stated that such an occurrence is rare. The radiologists at the Graduate Hospital have encountered only 1 patient in whom they demonstrated the presence of both lesions. The roentgen findings of achalasia were somewhat atypical and the esophagologist did not concur with the diagnosis.

Although the esophageal hiatus may be widened in patients with achalasia, it seems to us that such widening would be more difficult to demonstrate radiologically because of the very nature of the pathologic process. As a result of the relative stenosis which is almost invariably present in the distal esophagus in this condition, the quantity of contrast material passing through the esophageal hiatus is usually insufficient to distend the esophagus at this level to its maximum. For the same reason, there is usually not enough barium in the stomach to show a hiatal hernia.

According to Kay,[7] there is an interrelationship between the phrenoesophageal ligament and the inferior esophageal constrictor in the spincteric mechanism of the terminal esophagus. In patients with hiatal hernia this ligament is relaxed or atrophic, whereas in patients with achalasia it is taut. If this concept is correct, the two lesions rarely could coexist.

When surgical treatment is indicated for the patient with achalasia, our preference has been the Zaaiger modification of the Heller operation[13] in which only an anterior myotomy is carried out by way of an abdominal approach. The technical aspects of this operation have been described by Maingot.[9] In order to mobilize the lowermost part of the esophagus it is necessary to divide the peritoneum and phrenoesophageal ligament as they reflect from the diaphragm about the hiatus onto the esophagus. On occasion, strong retraction on the anterior margin of the hiatus is required for additional exposure of the narrowed segment of the esophagus.

With this in mind, it seems to us that the hiatal hernias occurring in these cases were the result of operative alteration of the hiatus or the disruption of the phrenoesophageal ligament. This resulted in reflux of gastric contents into the esophagus and esophagitis.

Most descriptions of modifications of the Heller operation have not included any hiatal reconstruction or gastric fixation as a requisite.[8, 9, 12] However, Wangensteen[11] has referred to this step in the description of the operation as he performs it, and thoracic surgeons have pointed out that one of the advantages of using the transthoracic approach in this operation is the ease with which the hiatus can be reconstructed.[3, 4] Ravitch,[10] in discussing a paper on achalasia, stated that the French surgeons, Santy, Lortat-Jacob, and D'Allaines, believe that the poor results after the Heller procedure are due to the presence of an unsuspected hiatal hernia, possibly produced by the operation, and that the hiatus should be tightened and the stomach fixed to the undersurface of the diaphragm in all cases. Sweet, in the same discussion, referred to his experience with a number of cases in which he repaired the hiatus at the time of esophagomyotomy, but he did not elaborate.[10] Baronofsky,[1] in a recent review on lesions of the esophagus, states that hiatal hernia is an inevitable occurrence with achalasia.

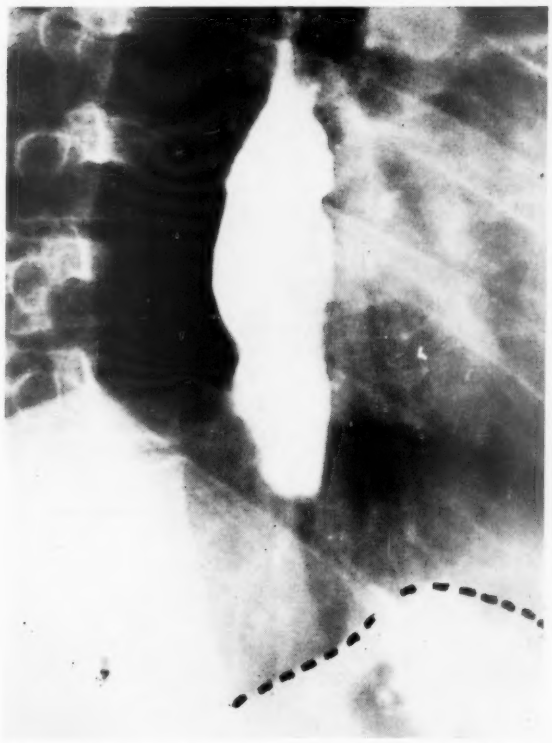
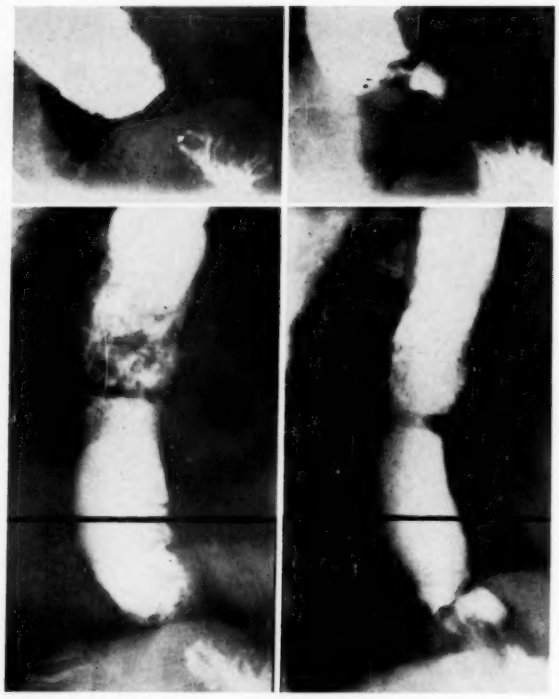


Fig. 7. Case 3. Initial roentgenogram which shows moderate esophageal dilatation to level of diaphragm due to achalasia.

Fig. 8. Case 3. Postoperative roentgenogram shows large hiatal hernia with gastric cardia extending well above diaphragm *(dotted lines)*.



A *B*

Fig. 9. Case 4. *A,* Preoperative roentgenogram which shows marked esophageal dilatation to level of diaphragm with marked stenosis below this level. Upper spot film of cardioesophageal juncture fails to demonstrate any hiatal hernia. *B,* Postoperative roentgenogram demonstrates no change in degree of dilatation. *Arrow* in upper spot film indicates herniated stomach above diaphragm *(dotted lines).*

A *B*

Fig. 10. Case 4. *A,* subsequent postoperative film study shows further dilatation of esophagus. *Arrow* indicates ulcer in herniated portion of stomach. *B,* Roentgenogram made after interposition operation. *Upper arrow* indicates interposed jejunal segment. *Lower arrow* indicates stomach below diaphragm.

In the light of our experience it is apparent that the incidence of hiatal hernia after the Heller operation is significant. Whether the hernia coexists with achalasia or is the result of the operative procedure is unimportant; however, meticulous attention to the restoration of the normal anatomic relationships of the hiatus is essential in the surgical treatment of achalasia. We would agree with Wangensteen,[11] Effler,[3] Ellis,[4] and others that, after esophagomyotomy, the hiatus should be narrowed to a normal diameter by placement of sutures in the edges of the right crux posterior to the esophagus, and that the severed phrenoesophageal ligament should be reconstructed so as to maintain the cardiac portion of the stomach below the diaphragm. If this step were included as an integral part of the operation, it seems to us that whether the approach were transthoracic or transabdominal would be of little importance.

SUMMARY

1. The coexistence of hiatal hernia and achalasia is probably rare.

2. The incidence of hiatal hernia as a complication of the Heller operation for achalasia, particularly when the abdominal approach is used, is significant. The symptoms produced by the complications of such hernias are usually more severe and distressing to the patient than those of the initial achalasia.

3. To avoid these complications, it is strongly urged that anatomic restoration of the esophageal hiatus and repair of the phrenoesophageal membrane be performed routinely in association with the Heller operation.

REFERENCES

1. Baronofsky, I. D.: Some recent advances in surgery of the esophagus, S. Clin. North America **39:** 1179-1192, 1959.
2. Brewer, M. S., Barnes, W. A., and Redo, S. F.: An evaluation of the operative procedure for achalasia, Ann. Surg. **144:** 823-828, 1956.
3. Effler, D. B.: Surgical treatment of achalasia

of esophagus and mega-esophagus *in* Current surgical management, Philadelphia, 1957, W. B. Saunders Company, pp. 230-236.

4. Ellis, F. H., Jr., Olsen, A. M., Holman, C. B., and Code, C. F.: Surgical treatment of cardiospasm, J. A. M. A. **166:** 29-36, 1958.
5. Euphrat, E. J., and Delmonico, J. E., Jr.: Coexistent achalasia of the esophagus and hiatal hernia of the cardiac end of the stomach, J. Thoracic Surg. **34:** 395-498, 1957.
6. Hawthorne, H. R., Frobese, A. S., and Nemir, P., Jr.: The surgical management of achalasia of the esophagus, Ann. Surg. **144:** 653-666, 1956.
7. Kay, E. B.: The inferior esophageal constriction in relation to lower esophageal disease, J. Thoracic Surg. **25:** 1-13, 1953.
8. Madden, J. L.: Atlas of technics in surgery, New York, 1958, Appleton-Century-Crofts, Inc., pp. 158-160.
9. Maingot, R.: Techniques in British surgery, Philadelphia, 1950, W. B. Saunders Company, pp. 358-373.
10. Ravitch, M. M.: Discussion of Tuttle et al.: Achalasia of esophagus: Further thoughts on surgical management, J. Thoracic Surg. **36:** 460-461, 1958.
11. Wangensteen, O. H.: Technique of achieving an adequate extramucosal myotomy in mega-esophagus (achalasia, cardiospasm, dystonia), Surg. Gynec. & Obst. **105:** 339-346, 1957.
12. Welch, C. E.: Surgery of the stomach and duodenum, Chicago, 1951, Year Book Publishers, Inc., p. 118.
13. Zaaiger, J. H.: Cardiospasm in aged, Ann. Surg. **77:** 615-617, 1923.

The toxicity of peritoneal fluid in strangulation obstruction: the influence of anesthesia

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The injection into normal animals of the peritoneal fluid from animals with strangulation obstruction has been used by many investigators to determine the toxicity of this fluid. The toxicity is undoubtedly augmented as a result of perforation of the strangulated bowel and spillage of bowel contents into the peritoneal cavity. Personal experience with various surgical preparations and study of the literature[7, 8] indicate that the short, closed-loop strangulation obstruction predisposes the bowel to perforation. When perforation occurs, death is due to peritonitis.

In an attempt to eliminate the complications associated with peritonitis, previous studies were restricted to peritoneal fluids from dogs in which the strangulated segment had not perforated.[10] Results from recent studies on rabbits with a nonperforated strangulated bowel indicated clostridial toxins to be the only major lethal factor in peritoneal fluid.[6] The contribution of the bacterial flora of this fluid to the lethality was slight.

However, Barnett and Hardy[5] have reported that the peritoneal fluid found late in the course of strangulation obstruction in dogs is lethal when injected into the peritoneal cavity of so-called "normal" dogs. In their control groups, an intraperitoneal injection of 3 ml. of peritoneal fluid per kilogram of body weight was a lethal dose for recipient dogs under intravenous pentobarbital sodium anesthesia. Barnett[2, 3] has concluded that the bacterial flora of peritoneal fluid are primarily responsible for the lethal properties of the canine fluid. This conclusion is at variance with that reached in this laboratory[6, 7, 9] and has prompted us to repeat and extend some of Barnett's work.

This is a report of the toxicity and bacteriologic properties of peritoneal fluid from dogs with a perforated bowel due to closed-loop strangulation obstruction. The dominant influence of anesthesia on the outcome of toxicity studies is documented.

METHODS

Eleven adult mongrel dogs were maintained on Purina laboratory chow for 2 weeks prior to operation. A closed-loop strangulation obstruction was established under intravenous pentobarbital sodium anesthesia, with all the aseptic precautions used in a hospital operating room. The length of the small intestine was measured. At a point 75 per cent of the distance distal to the ligament of Treitz, the vein from a 10 cm. segment of small intestine was ligated, and a closed-loop obstruction was created by tightly tying each end of the segment with umbilical tape (Fig. 1). The abdomen was closed and the animals were returned to cages for constant observation.

Autopsy was performed immediately upon death of the animals. Reddish-black, foul-smelling fluid was aspirated from the peri-

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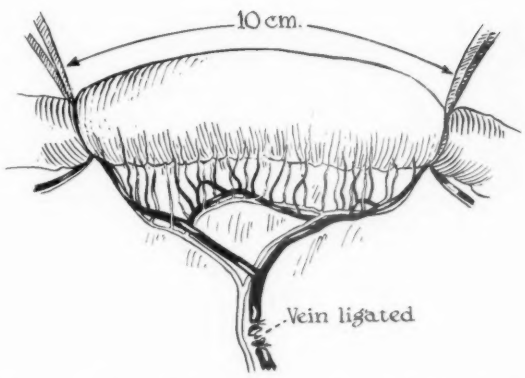

Fig. 1. The technique for creating a short, closed-loop intestinal strangulation obstruction in the dog.

toneal cavity. Each preparation was noted as "perforated" or "nonperforated." The peritoneal fluid from each dog was refrigerated for not more than 24 hours before being pooled with that collected from other dogs which had died on the same day with similarly perforated or nonperforated bowels. Pooled peritoneal fluid was occasionally used immediately after pooling. Otherwise, the pools were frozen and thawed immediately before use.

Recipient dogs, which had been maintained on Purina laboratory chow for 2 weeks, were given an intraperitoneal injection of pooled peritoneal fluid. The abdomen was prepared in routine surgical fashion. A 14 gauge needle was inserted into the mid-abdomen on the right and air injected and aspirated to assure that the needle was not inserted into the gastrointestinal tract. An 8-inch sterile polyethylene tube was inserted through the needle and the needle was removed. The peritoneal fluid was injected through the polyethylene tube. Despite occasional vomiting or diarrhea after the injection, death was the criterion for toxicity.

One group of 7 recipient dogs were anesthetized intravenously with pentobarbital sodium before the intraperitoneal injection to duplicate the condition employed by Barnett and Hardy.[5] The maximum dosage administered was 0.8 ml. per kilogram of a 3 per cent solution of pentobarbital sodium.

No pentobarbital was administered to another group of 11 recipient dogs. Neither the preliminary manipulations nor the intraperitoneal injections appeared to cause immediate discomfort to the animals.

When peritoneal fluids were pooled, specimens were taken for quantitative bacteriologic analysis according to the protocol employed in this laboratory.[6]

Peritoneal fluid was sterilized by filtration after being centrifuged at 24,000 g for 30 minutes at 0° C. The supernatant was then passed through a No. 142 Millipore laboratory sterilizing filter. Aerobic and anaerobic cultures of this filtrate were sterile.

RESULTS

The 11 animals with strangulation obstruction had a mean survival time of 26 hours (range 18 to 39 hours), and produced a mean volume of 190 ml. of peritoneal fluid (range 12 to 650 ml.). The closed-loop of 7 of these dogs (64 per cent) was collapsed, hemorrhagic, necrotic, and visibly perforated or disintegrated (Fig. 2). The remaining 4 animals showed distended, necrotic, and hemorrhagic loops which were not visibly perforated. To inflict the maximal insult to recipient animals, only fluids pooled from animals with a perforated loop were injected into the peritoneal cavity.

Table I shows the effect of intravenous pentobarbital anesthesia upon the lethality of pooled peritoneal fluid for recipient dogs. These animals recovered from anesthesia within 6 hours. Intraperitoneal injection of

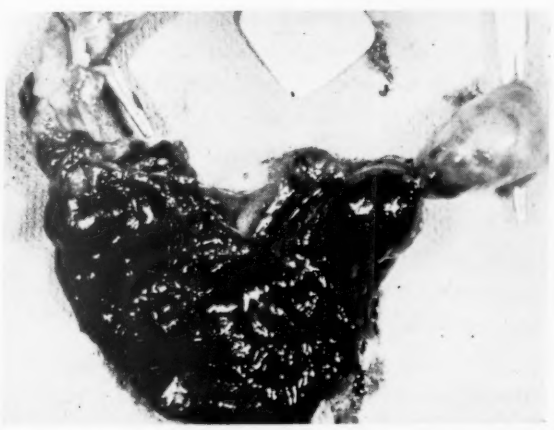

Fig. 2. A typical perforated closed-loop strangulation obstruction at autopsy in a dog.

Table I. *Effect of intravenous pentobarbital sodium anesthesia on the response of dogs injected intraperitoneally with pooled peritoneal fluid**

Condition	*Dosage of peritoneal fluid injected (ml./kg.)*	*No. of dogs† injected with peritoneal fluid*	*No. of dogs surviving*
Anesthesia	3	5	0‡
No anesthesia	3	7	7
	6	2	2
	9	2	2

*Pooled peritoneal fluid taken from dogs with perforated strangulated loop.

†Mean weight 9.5 kilograms (range 7.0 to 13.0 kilograms).

‡Death in 5, 10, <16, <16, and 16 hours.

fluid at 3 ml. per kilogram of body weight was uniformly lethal within 24 hours in recipient dogs which had been anesthetized initially. This same dosage was uniformly nonlethal when recipient dogs had not been anesthetized.

Anesthesia was omitted in other studies in an attempt to establish a lethal dosage in unanesthetized animals. Dogs challenged with dosages of 6 ml. per kilogram of body weight and 9 ml. per kilogram of body weight all survived. Thus, pooled peritoneal fluid is essentially nonlethal for the unanesthetized dog. Conversely, pooled peritoneal fluid is lethal for the anesthetized dog. These data indicate that anesthesia renders the recipient dog more than 3 times as sensitive as the unanesthetized dog to the peritoneal fluid.

Table II summarizes the bacteriologic properties of pooled peritoneal fluids. Each pool was composed of fluid from 2 or 3 donor dogs. There appears to be no great degree of variation in the results of the quantitative bacteriologic studies of any pool. The low bacterial counts suggest that no single species contributed to an overwhelming infection.

A sterilized pool of peritoneal fluid was investigated to determine whether or not the removal of bacteria from the fluid would alter its uniform lethality for anesthetized recipient dogs. Accordingly, 2 anesthetized dogs were challenged with sterile peritoneal fluid (3 ml. per kilogram of body weight) given intraperitoneally. Both dogs survived.

If the dogs which received the intraperitoneal injections of peritoneal fluid were dying solely from bacterial infection, as has been suggested, then, comparison of the counts of the injected fluid with counts of fluid obtained at autopsy from the same animals might show significant increases in bacteria indicative of the most important organism or organisms. Table III presents data concerning 3 of the 5 anesthetized recipient dogs that died during observation. The animal which died at 5 hours had not completely recovered from anesthesia. The 2 other dogs had slight to mild degrees of generalized peritonitis. The volume of peritoneal fluid collected at death was approximately twice that injected. This fluid was lighter in color and less foul-smelling than the material with which the animal had been challenged initially. Only the fluid obtained from the animal which survived the longest was malodorous. The quantitative bacteriologic studies indicated that there was no increase in the number of bacteria present in the peritoneal cavity. Rather, the total number of organisms in the peritoneal fluid was *less* than that in the fluids with which the dogs had been challenged.

All unanesthetized recipient dogs were sacrificed 1 week after challenge. At autopsy, the abdominal findings were negative and there was no indication of peritonitis.

DISCUSSION

The most provocative finding of this study is that the unanesthetized dog survives challenge with a peritoneal fluid that is lethal for the anesthetized dog. The unanesthetized dog survives challenge with 3 times the dosage of peritoneal fluid needed to kill the anesthetized dog. Indeed, it would have been concluded that peritoneal fluid from dogs with strangulation obstruction with perforation was *nonlethal* at the dosages assayed if no anesthesia had been used.

There are recent reports of experiments in which peritoneal fluid from dogs with un-

treated closed-loop strangulation obstruction was injected into anesthetized dogs. The results were compared with the results of experiments in which peritoneal fluid from antibiotic-treated animals was injected into unanesthetized dogs.[1, 4] Barnett and Hardy[5] emphasized in their initial study that intraperitoneal injection caused such severe discomfort to test animals that it was humane to anesthetize them. However, a more recent report[2] states: *"It was not necessary to anesthetize recipient animals because they registered no discomfort after injection of fluid into the peritoneal cavity"* (italics ours) when there had been no evidence of gross rupture or odor of untreated gangrenous bowel. To make the studies truly comparable, anesthesia should have been administered to, or withheld from, *all* animals challenged with peritoneal fluid.

Studies of closed-loop strangulation obstruction in rabbits have provided evidence that clostridial exotoxins were the major lethal component in peritoneal fluid, and that the bacterial flora per se were of slight importance.[6] It was therefore of considerable interest to observe that sterile peritoneal fluid from a similar preparation in the dog was not lethal for anesthetized dogs. Barnett[2, 3] has also found that canine peritoneal fluid sterilized by filtration is not lethal for anesthetized dogs, and has concluded that bacterial cells were essential for a lethal outcome. As raw peritoneal fluid is not lethal for the unan-

Table II. *Results of bacteriologic studies of pooled peritoneal fluid from dogs with perforated strangulated loop*

Pool No.	No. of bacteria per milliliter of peritoneal fluid				
	Clostridia	*Bacteroides*	*Coliforms*	*Streptococci*	*Total*
1	9×10^5	46×10^5	15×10^3	12×10^6	17.4×10^6
3	4×10^5	4×10^5	3×10^5	31×10^3	1.1×10^6
4	16×10^5	35×10^5	16×10^5	30×10^6	36.7×10^6
Mean	9.7×10^5	28×10^5	6.2×10^5	1.4×10^6	18.4×10^6

Table III. *Comparison of results of bacteriologic studies and descriptions of peritoneal fluid injected intraperitoneally into anesthetized dogs and the fluid aspirated from the peritoneal cavity of the same dogs at death*

	Volume of fluid (ml.)	No. of bacteria per milliliter of peritoneal fluid					*Total No. of bacteria*	*Blood culture at death*	*Remarks on peritoneal fluid at death and autopsy report*
		Clostridia	*Bacteroides*	*Coliforms*	*Streptococci*	*Aerobic gram-positive rods*			
Dog No. 5308 (5 hr.)*									
Injected	30	4×10^3	18×10^3	440	60	10×10^5	306×10^5	Negative	Red, cloudy, clotted fluid
Recovered	60	None	None	380	12×10^4	None	72×10^5		
Dog No. 5411 (10 hr.)*									
Injected	25	9×10^5	46×10^5	15×10^3	112×10^6	None	443×10^6	*E. coli*	Sanguinopurulent, malodorous fluid; generalized, moderately severe peritonitis
Recovered	50	150	None	500	2×10^6	2×10^6	200×10^6		
Dog. No. 5408 (16 hr.)*									
Injected	25	9×10^5	46×10^5	15×10^3	112×10^6	None	443×10^6	Clostrida	Bloody, malodorous fluid; generalized mild peritonitis
Recovered	25	2×10^3	None	20	23×10^4	32×10^4	14×10^6		

*Survival time.

esthetized dog, the bacterial flora of raw peritoneal fluid may be essential for a lethal outcome only when the recipient dog has been anesthetized at the time of injection. The evidence presented does not support the conclusion that bacteria themselves are primarily responsible for death in strangulation obstruction. The problem warrants continued investigation.

Moreover, a relatively low bacterial count was found in peritoneal fluids from dogs with closed-loop strangulation obstruction and perforation. Additional evidence for the lack of importance of the bacteria per se is found in the results of bacteriologic studies of the peritoneal fluid of anesthetized recipient dogs, which reveal a significant decrease in the total number of bacteria as well as significant diminution of clostridial, bacteroides, and coliform groups. The lethal effect of the peritoneal fluid in the anesthetized dog, in contrast to its lack of effect in the unanesthetized animal, suggests that the anesthetized dog's defense mechanisms are altered to the extent of being incapable of overcoming the stress.

The superior tolerance of the unanesthetized animals to this stress is further demonstrated by their normal condition at the time of sacrifice 1 week after challenge.

SUMMARY

1. Peritoneal fluid from dogs with experimental strangulation obstruction was not lethal when injected intraperitoneally into unanesthetized dogs.

2. This peritoneal fluid was lethal for dogs anesthetized with intravenous pentobarbital sodium, but was not lethal when it had been sterilized by filtration.

3. Unanesthetized recipient dogs survived challenge with 3 times the dosage of peritoneal fluid needed to kill the anesthetized dogs.

4. The bacterial counts of peritoneal fluid from dogs with closed-loop strangulation obstruction were low and did not define any single bacterial species as the cause of an overwhelming infection.

5. The role of bacteria in death from strangulation obstruction warrants further investigation.

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REFERENCES

1. Barnett, W. O.: The efficacy of Chloromycetin in the treatment of strangulation obstruction, Ann. Surg. **149:** 471, 1959.
2. Barnett, W. O.: Experimental strangulated intestinal obstruction—a review, Gastroenterology **39:** 34, 1960.
3. Barnett, W. O.: Strangulation obstruction: pathophysiology and management, Mississippi Doctor **36:** 72, 1958.
4. Barnett, W. O., and Doyle, R. S.: The effects of neomycin upon the toxicity of peritoneal fluid resulting from strangulation obstruction, SURGERY **44:** 442, 1958.
5. Barnett, W. O., and Hardy, J. D.: Observations concerning the peritoneal fluid in experimental strangulated intestinal obstruction: the effects of removal from the peritoneal cavity, SURGERY **43:** 440, 1958.
6. Bornside, G. H., and Cohn, I., Jr.: Clostridial toxins in strangulation intestinal obstruction in the rabbit, Ann. Surg. **152:** 330, 1960.
7. Cohn, I., Jr.: Strangulation obstruction, Springfield, 1961, Charles C Thomas, Publisher.
8. Cohn, I., Jr.: Strangulation obstruction, Surg. Gynec. & Obst. **103:** 105, 1956.
9. Cohn, I., Jr., and Hawthorne, H. R.: The role of *Clostridium welchii* in strangulation obstruction, Ann. Surg. **134:** 999, 1951.
10. Nemir, P., Jr., Hawthorne, H. R., Cohn, I., Jr., and Drabkin, D. L.: The cause of death in strangulation obstruction. I. Clinical course, chemical, bacteriologic, and spectrophotometric studies. II. Lethal action of the peritoneal fluid, Ann. Surg. **130:** 857, 874, 1949.

Experimental arterialization of the liver

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The development of a preparation in which the arterial blood supply to the liver is augmented has attracted attention for a variety of reasons. It has been the opinion that acceleration of liver regeneration and improvement of hepatocellular function might result from arterialization of the liver. Also, increasing the flow of oxygenated blood to the liver has been proposed to avoid the immediate deterioration of liver function associated with portacaval anastomosis for cirrhosis with portal hypertension.

Several techniques have been used in an effort to make the portal vein an arterial conduit. In 1916, Narath[13] anastomosed the left renal artery to the splenic vein. The procedure was abandoned by the author himself and replaced by anastomosis between the hepatic artery and portal vein. This was subsequently studied by Schilling, McKee, and Wilt,[17] and Bariatti, Servello, and Taccani.[1] Paterni[14] and Servello and Rossi[19] investigated splenic arteriovenous anastomosis after removal of the spleen. Direct anastomosis of the splenic artery into the portal vein was also studied. Servello and Petronio[18] favored anastomosis of the aorta to the splenic vein as a side-to-side preparation after splenectomy.

Finally, several investigators[2, 3, 9, 11, 12] have performed anastomoses between the aorta and portal vein by means of a venous autograft or prosthetic graft. We have used this operation, performed in combination with an end-to-side portacaval shunt to detour portal venous blood away from the liver, as a means of studying the effects of total arterialization of the liver. The morphologic and biochemical changes associated with the conversion of the portal vein and its branches into an arterial system constitute the basis of this presentation.

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*John and Mary R. Markle Scholar.

METHODS

Adult mongrel dogs were anesthetized intravenously with sodium pentobarbital and ventilated by means of a positive-pressure respirator. Before operation, peripheral venous blood was drawn and serum protein, albumin, globulin and alkaline phosphatase levels, thymol turbidity, prothrombin time, and icterus index were determined. With the right side of the animal elevated at approximately 45 degrees, a thoracoabdominal incision was made through the tenth intercostal space. The pleural and peritoneal cavities were entered and then joined by means of transection of the diaphragm in the direction of the incision. Initial liver biopsies were carried out in several of the animals.

The portal vein was dissected free from surrounding tissues and the cranial (superior) mesenteric artery was occluded with a bulldog clamp. Atraumatic vascular clamps were then applied to the splanchnic and hepatic ends of the dissected portal vein, and the portal vein was transected close to the liver. An end-to-side portacaval anastomosis was performed between the splanchnic end of the

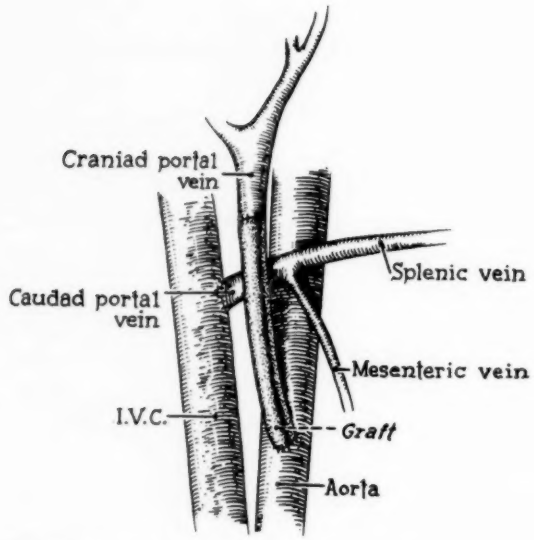

Fig. 1. Diagram of operative preparation.

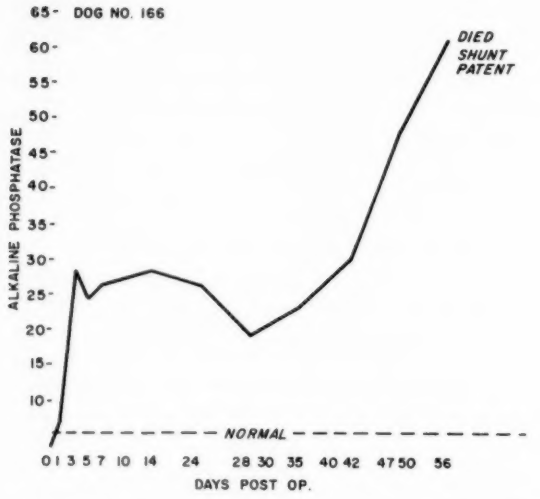

Fig. 2. Alkaline phosphatase levels in dog with persistently patent arterial shunt.

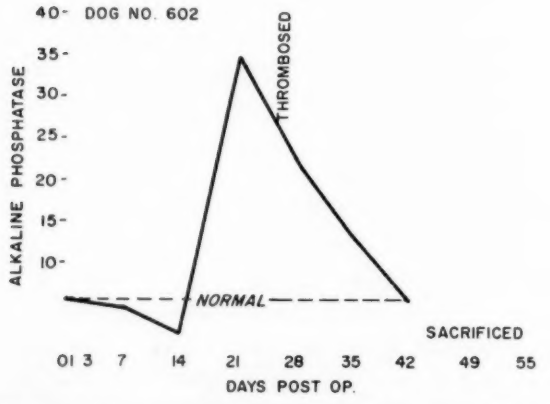

Fig. 3. Alkaline phosphatase levels in dog with thrombosed shunt.

portal vein and the side of the inferior vena cava. The mesenteric arterial clamp was then removed. A prosthetic graft, with an internal diameter of 6 mm. approximating the lumen of the portal vein, was anastomosed end-to-end to the hepatic end of the portal vein. An end-to-side anastomosis was then effected between the remaining end of the prosthetic graft and the side of the aorta below the renal arteries (Fig. 1). The wounds were closed in layers and suction was applied to the right pleural space for several hours.

Liver function studies were performed every other day during the first postoperative week and then at weekly intervals until the dog died or was sacrificed. At varying intervals, liver biopsies were made, either by the closed technique with a Vim-Silverman needle or, more frequently, by open biopsy during laparotomy. The patency of the graft was determined by palpation at each laparotomy. All animals were autopsied, and the liver was removed with the portal vein and graft attached so that the final status of the graft and liver could be evaluated.

RESULTS

The procedure of portacaval anastomosis and arterialization of the liver was performed in 25 dogs. Thirteen of the animals died during the immediate postoperative period. Hemorrhage, intestinal infarction, and massive atelectasis were the most common causes contributing to failure in these cases. In some dogs, however, a specific cause of death was not apparent. Two animals died several weeks after operation without sufficient study and could not be fully evaluated.

An investigation of the results and findings in the remaining 12 dogs constitutes the basis of this report. In all of these, both the portacaval and aortic-portal anastomoses were patent at the conclusion of the operation. Liver function studies performed during the postoperative period in these animals showed essentially no alteration in total protein, albumin, or globulin levels, thymol turbidity, prothrombin time, or icterus index.

A significant alteration in the serum alkaline phosphatase level was noted and a

relationship between this level and the patency of the aortic-portal graft was demonstrated (Table I).

In all but 1 of the dogs (No. 59-230) in which successful arterialization of the liver was accomplished, the maintenance of a patent arterial graft was accompanied by a steadily rising alkaline phosphatase level. Fig. 2 represents the typical rise in alkaline phosphatase in a dog with a persistently patent arterial shunt. This finding was contrasted with the change that occurred when the graft thrombosed late in the postoperative course. A consistent decrease in the serum alkaline phosphatase level occurred subsequent to the thrombosis (Fig. 3).

The fate of each dog is outlined in Table II. Six animals were sacrificed after postoperative periods varying between 2 weeks and 4 months, with a mean survival time of 10 weeks. Five of the dogs demonstrated thrombosis of the graft and absence of flow through the hepatic end of the portal vein at the time of sacrifice. In 3 dogs, which

Table I. *Relationship of level of serum alkaline phosphatase* to patency of graft in total arterialization of the liver*

	Alkaline phosphatase levels			
Dog No.	*Preoperative*	*After shunt (maximum)*	*Immediately before death or sacrifice*	*Final graft status*
58-314	1.5	46.0	9.3*	Thrombosed
58-426	2.0	35.0	35.0	Patent
58-476	2.2	18.0	18.0	Patent
58-554	2.0	31.2	31.2	Patent
58-555	1.5	46.3	46.3	Patent
58-559	2.5	52.5	52.5	Patent
58-602	5.5	35.0	5.5	Thrombosed
59-125	1.0	10.0	4.5	Thrombosed
59-131	4.0	26.0	7.5	Thrombosed
59-138	2.0	9.5	2.5	Thrombosed
59-166	3.5	61.6	61.6	Patent
59-230	0.5	8.0	1.5	Patent

*Measured in Bodansky units. Normal range in 24 healthy dogs was 0.5 to 5.5 B.U., with an average value of 2.2 B.U. and a mean of 2.0 B.U.

Table II. *Correlation between survival, status of graft, and alkaline phosphatase*

Dog No.	*Graft*	*Final alkaline phosphatase level*	*Results*
58-314	Thrombosed	9.3	Survived
58-426	Patent	35.0	Died 87 days postoperatively
58-476	Patent	18.0	Died 8 days postoperatively (after aortogram)
58-554	Patent	31.2	Died 52 days postoperatively (after aortogram)
58-555	Patent	46.3	Died 57 days postoperatively
58-559	Patent	52.5	Died 42 days postoperatively (after liver biopsy)
58-602	Thrombosed	5.5	Survived
59-125	Thrombosed	4.5	Survived
59-131	Thrombosed	7.5	Survived
59-138	Thrombosed	2.5	Survived
59-166	Patent	61.6	Died 29 days postoperatively (after liver biopsy)
59-230	Patent	1.5	Survived

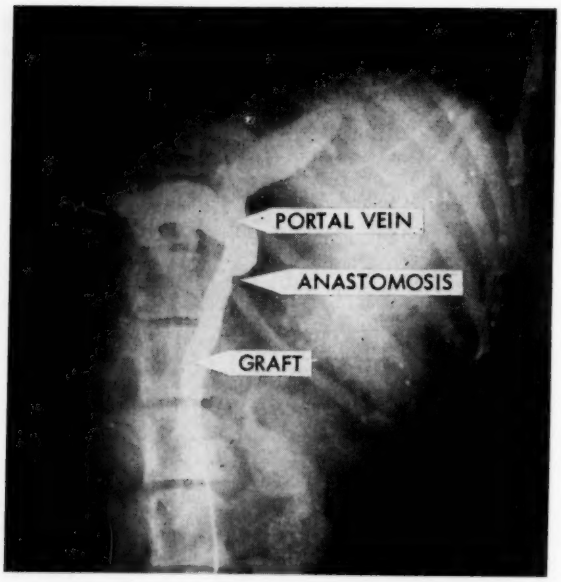

Fig. 4. Arteriogram of patent arterial shunt.

Fig. 5. Photomicrograph of portal vein after arterialization.

were sacrificed after 1 month, an arterial flow to the liver via the portal vein had been demonstrated at laparotomy 2 to 3 weeks after the initial procedure. Thus, in these 3 animals, thrombosis of the graft occurred late in the postoperative course. In all 5 dogs the alkaline phosphatase level immediately prior to death was normal. The sixth dog (No. 59-230) in the survival group had a patent shunt at the time of sacrifice and this is the one instance in which a normal alkaline phosphatase level was associated with a patent arterial shunt.

Six animals died 1 week to 2 months after operation. All had patent arterial shunts demonstrated at autopsy and an elevated alkaline phosphatase level in the serum drawn shortly before death. Two of the animals died of hemorrhage from a biopsy site in the liver. One biopsy had been performed as a closed procedure with the Vim-Silverman needle and the other had been carried out during laparotomy. Two dogs died of hemorrhage associated with aortography which had been performed to demonstrate the patency of the graft. The remaining 2 animals died of undetermined causes.

Arterialization of the portal venous system resulted in consistent dilatation of the portal vein and its major branches. This was apparent on an arteriogram performed 2 months after the operative procedure (Fig. 4) and also at autopsy of all animals with patent grafts. In addition to the dilatation of the lumen, a thickening of the wall of the portal vein was apparent (Fig. 5).

The changes in the liver associated with arterialization were inconsistent. The findings varied from a grossly and microscopically normal liver to a liver which was grossly nodular (Fig. 6) and histologically consistent with cirrhosis. In this instance, the classic components of cirrhosis, including deterioration of liver cells, proliferation of bile ductules, and increased periportal fibrous tissue, were all present (Fig. 7).

DISCUSSION

Dogs which survived for long periods with splanchnic blood bypassing the liver and with a patent arterial shunt consistently showed deterioration of their general condition. Most dogs maintain or gain weight on our regular kennel diet, but the animals in this series lost considerable amounts of weight. The fact that the livers had been arterialized was demonstrated by the 2 fatal arterial hemorrhages which occurred as a direct result of liver biopsy.

There has been disagreement concerning the effects of arterialization on the portal vein itself and on the hepatic cells. Our studies demonstrated a thickening and dilatation of the vein with inconsistent findings in

the liver itself which ranged from a normal gross and microscopic picture to one suggestive of portal cirrhosis. Bariatti, Servello, and Taccani[1] found no changes when the hepatic or right renal artery was implanted into the portal vein. Schilling, McKee, and Wilt[17] demonstrated aneurysmal dilatation of the portal vein at the site of anastomosis to the hepatic artery. Using a technique similar to ours, Rather and Cohn[16] found the main portal vein to be much greater in caliber and thickness than in normal dogs. They also noted thrombus formation in the portal vein and acute necrotizing vasculitis with secondary constriction in the portal radicals of most dogs. Fisher and his associates,[4] however, found no intrahepatic damage. McCredie's group[11] had a similar experience. In the 1 reported case of a patient with an established hepatoportal arterial venous fistula and secondary partial arterialization of the liver, thrombi were noted in many of the portal vein tributaries in the hilum of the liver.[20]

The most interesting finding in our investigation was the correlation between elevation of serum alkaline phosphatase level and patency of arterial shunt. Gutman[5] pointed out that in the dog, an elevation of the serum alkaline phosphatase level occurs when the patency of the intra- or extrahepatic biliary tract is minimally compromised. It was his opinion that this rise was caused by obstruction of biliary flow, rather than by hepatocellular damage. It is conceivable that, in our experiment, increased flow through the portal bed within the liver impinged upon the passage of bile through the interhepatic ducts. Evidence favoring this interpretation is the fall in phosphatase level which accompanied thrombosis of the arterial shunt. Tests of hepatocellular function, such as thymol turbidity, prothrombin time, and serum protein determinations, gave no evidence of hepatocellular damage in the majority of our animals.

In 1952, Hunt[8] treated 4 patients with advanced liver disease by means of a portacaval shunt and anastomosis of the right

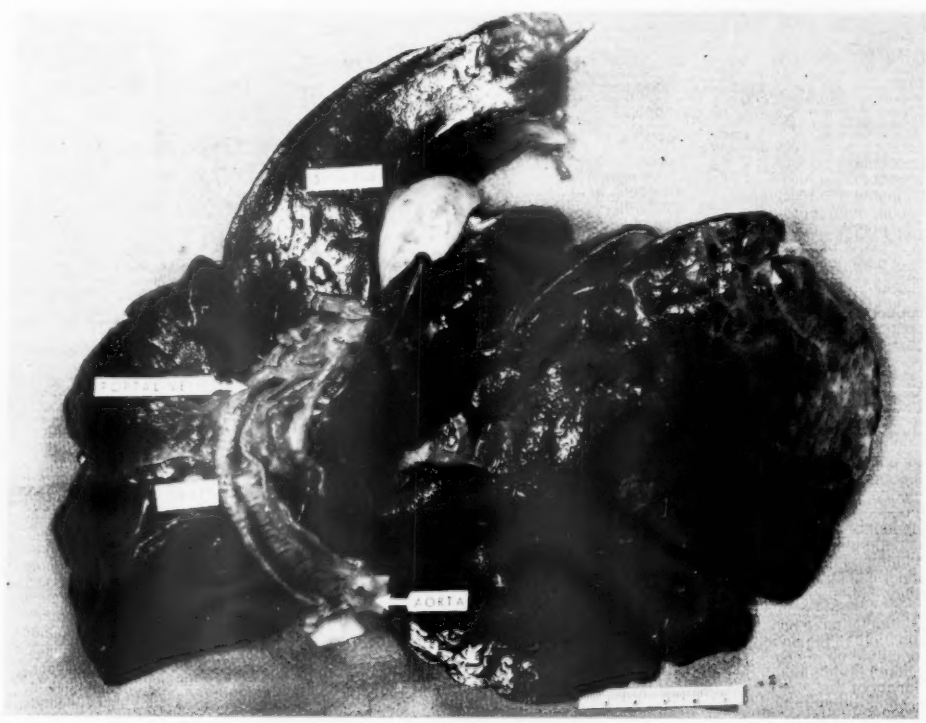

Fig. 6. Gross specimen which demonstrates nodularity of liver and patent arterial graft.

Fig. 7. Photomicrograph of liver (from specimen shown in Fig. 6).

renal artery to the hepatic end of the portal vein. All patients died within a few days. Jones and his co-authors[10] implanted the right gastroepiploic artery into the stump of the portal vein after portacaval anastomosis in 1 patient. No improvement in liver function was noted. Several authors[15, 21, 22] have reported occasional cases in which the splenic artery and vein were anastomosed to arterialize the liver, with suggestion of favorable results. McCredie and his associates[11] showed that in dogs rendered cirrhotic with carbon tetrachloride, portacaval anastomosis was tolerated better alone than in combination with liver arterialization.

Herrick[7] demonstrated that when the hepatic artery was perfused in the cirrhotic liver, the primary return was by way of the portal vein. Hales and his co-authors,[6] using injection and corrosion studies of cirrhotic livers, indicated that the most consistent findings were enlargement of the hepatic artery and increase in the injectable hepatic arterial bed accompanied by a reduction in the hepatic and portal venous systems. These authors believed that arterialization of the portal vein would deprive a cirrhotic liver of its primary outflow and result in an overall decrease rather than an increase in hepatic arterial flow. Our experiments, performed on the noncirrhotic dog, demonstrated generalized deterioration in the animal's condition and marked rise in the level of the serum alkaline phosphatase. These findings support the concept that total arterialization of the liver is not a beneficial procedure.

SUMMARY

Total arterialization of the liver in the normal dog was accomplished by construction of an end-to-side portacaval shunt and connection of the aorta to the hepatic end of the portal vein by means of a prosthetic graft. In the dogs that survived this procedure, a correlation was demonstrated between the patency of the arterial shunt and the level of serum alkaline phosphatase. Thickening and dilatation of the portal vein were seen at autopsy, but no consistent alteration of liver structure was observed in association with arterialization.

REFERENCES

1. Bariatti, R., Servello, M., and Taccani, C.: Creazione di anastomosi artero-venosa epatico-portale, Arch. ed. atti d. Soc. ital. di chir. **52:** 62, 1950.
2. Cohn, R., and Herrod, C.: Some effects upon the liver of complete arterialization of its blood supply, SURGERY **32:** 214, 1952.
3. Fisher, B., Russ, C., and Updegraff, H.: A suitable technique for the total arterialization of the dog liver, SURGERY **35:** 879, 1954.
4. Fisher, B., Russ, C., Fedor, E., Wilde, R., Engstrom, P., and Fisher, E. R.: Further experimental observations on animals with arterialized livers, SURGERY **38:** 181, 1955.
5. Gutman, A. B.: Serum alkaline phosphatase activity in diseases of the skeletal and hepatobiliary systems, Am. J. Med. **27:** 875, 1959.
6. Hales, M. R., Allan, J. S., and Hall, E. M.: Injection-corrosion studies of normal and cirrhotic livers, Am. J. Path. **35:** 909, 1959.
7. Herrick, F. C.: An experimental study into the cause of the increased portal pressure in portal cirrhosis, J. Exper. Med. **9:** 93, 1907.
8. Hunt, A. M.: The surgical treatment of Banti's syndrome, Brit. M. J. **2:** 4, 1952.
9. Jamison, W. L., Dugan, T. J., Bailey, C. P., and Ramirez, H. P. R.: Experiences in experimental porto-caval anastomosis, SURGERY **30:** 484, 1951.
10. Jones, S. A., Reynolds, T. B., Schultz, E. B., and Gregory, G.: Arterialization of the human liver following portacaval anastomosis, West. J. Surg. **63:** 574, 1955.
11. McCredie, J. A., Doggart, J. R., and Welbourn, R. B.: Total arterialization of the liver, Brit. J. Surg. **45:** 83, 1958.

12. Markowitz, J., Rappaport, A., and Scott, A. C.: Prevention of liver necrosis following ligation of the hepatic artery, Proc. Soc. Exper. Biol. & Med. **70:** 305, 1949.
13. Narath, V.: Über Enstehung des anämische Lebernekrose nach Unterbindung der arteria hepatica und ihre Verhütung durch arteroportal anastomose, Deutsch. Ztschr. Chir. **135:** 305, 1916.
14. Paterni, L.: Disordine circolatorio del fegato nelle cirrosi. Orientamenti in terapia chirurgica, Policlinico (sez. prat.) **56:** 253, 1949.
15. Pettinari, V.: Iperpressione portale e suo trattamento chirurgico. Read before the Societa Medica di Verona, February 15, 1955.
16. Pettinari, V.: Ipersplenismo, iperpressione portal e splenopatie cirrogene, Chir. e pat. Sper. **2:** 4, 1954.
17. Pettinari, V.: Moderni indirizzi e realizzazioni nella terapia chirurgica della cirrosi epatica, Boll. Soc. Piemontese Chirurgica **22:** 2, 1952.
18. Rather, L. J., and Cohn, R.: Some effects upon the liver of complete arterialization of its blood supply. III. Acute vascular necrosis, SURGERY **34:** 207, 1953.
19. Schilling, J. A., McKee, F. W., and Wilt, W.: Experimental hepatic-portal arterio-venous anastomoses, Surg. Gynec. & Obst. **90:** 473, 1950.
20. Servello, M., and Petronio, R.: Some experimental methods for arterialization of the liver: the aortosplenic anastomosis, J. Internat. Coll. Surgeons **25:** 448, 1956.
21. Servello, M., and Rossi, R.: L'arterializzazione del fegato mediante anastomosi artero-venose spleno-spleniche e spleno-portali, Arch. ital. chir. **76:** 300-320, 1953.
22. Servello, M., and Rossi, R.: Studio istochimico sul comportamento del glicogeno, dei lipidi, delle fosfatasi alcaline e degli acidi nucleinici epatici dopo arterializzazione della porta mediante anastomosi artero-venosa, Pat. Sper. e cir. **1:** 352, 1953.
23. Strickler, J. H., Lufkin, H., and Rice, C. O.: Hepatic-portal arterio-venous fistula; a case report, SURGERY **31:** 583, 1952.
24. Trivellini, A.: Problemi di chirurgia nell'pertensione portale: L'arterializzazione portale, Arch. ed. atti d. Soc. ital. di chir. **56:** 66, 1954.
25. Valdoni, P.: *In* Lanzara: Ipertensione portale, Recenti progr. med. **12:** 191, 1952.

Experimental production of esophageal varices in the dog

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The experimental production of esophageal varices in animals has been the target of several investigators. Such a preparation would offer obvious opportunities for study. Laufman and co-workers[2] have succeeded in producing true submucosal varicosities in the *Macaca mulatta*. These appear to resemble the varicosities seen in patients with portal hypertension. This technique was based on the observation of McKee and his associates,[3] who, in 1949, reported the production of ascites and portal hypertension in dogs in which the supradiaphragmatic vena cava had been partially occluded. Milnes and Child[4] noted that monkeys and human beings could survive sudden portal vein occlusion, but with dogs this maneuver almost invariably resulted in death. Because the monkey has several disadvantages as an experimental subject, an attempt was made to produce esophageal varices in the dog.

In the past, numerous attempts have been made to develop a canine preparation with esophageal varices. In 1939, Rousselot and Thompson[5] attempted to produce congestive splenomegaly by the injection of silicone particles into the splenic vein of dogs. However, they were unsuccessful in producing esophageal varices.

Baronofsky and Wangensteen,[1] in 1945, produced esophageal varices by the gradual occlusion of the portal and splenic veins of dogs. They apparently were able to produce hemorrhage by administration of a histamine-beeswax preparation.

Tamiya and Thal[6] demonstrated that large esophageal varices could be produced in the dog by a two-stage operation. The first stage involved resection of spleen, omentum, and tail of pancreas, and thereby the removal of potential collateral pathways. An arteriovenous shunt was created between splenic artery and left gastric vein. The second stage consisted of partial occlusion of the splenic vein. The disadvantages of the procedure were twofold. First, the technique depended on arteriolization of the portal system for its success, and second, a high failure rate was associated with closure of the arteriovenous shunt. These authors[7] also were able to produce varices in the dog by a two-stage technique which consisted primarily of an aorto-portal shunt followed in 3 to 4 weeks by gradual occlusion of the portal vein on the hepatic side of the shunt. When a large shunt of this type was employed it was unnecessary to remove collateral venous circulation in order to produce gastroesophageal varices.

Laufman[2] mentions that he has noted small esophageal varices in some dogs with band-induced portal hypertension. It is interesting to note that the varices in his animals failed to bulge into the lumen, a fact which suggests that they did not lie in a submucosal location.

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The present report concerns an attempt to produce esophageal varices in the mongrel dog by a staged venous ligation technique similar to that recently reported by Laufman for the *Macaca rhesus*. The first stage consisted of azygos vein interruption and banding of the supradiaphragmatic vena cava to achieve a 50 per cent decrease in its circumference. This was followed by the development of ascites and portal hypertension over a 4 to 6 week period. When ascites was well established, the second stage, consisting of portal vein ligation or constriction, was carried out. Splenoportograms were performed 2 to 4 weeks later to demonstrate the presence of esophageal varices. Autopsy studies and esophagoscopy were also carried out to confirm the presence and exact location of the varices.

METHODS

Forty mongrel dogs, weighing between 5 and 20 kilograms, were subjected to either two- or three-stage procedures designed to open up pathways of potential collateral venous flow from the portal system in an effort to experimentally establish esophageal varices.

All operations were conducted under sterile conditions, with intravenously administered pentobarbital sodium, 25 mg. per kilogram, as the anesthetic agent. Endotracheal intubation and a positive-pressure respirator were routinely employed. The staged procedures consisted of (1) right thoracotomy with banding of the supradiaphragmatic inferior vena cava to produce a 50 per cent constriction, (2) ligation of the azygos vein below the superior intercostal vein, and (3) celiotomy with subcutaneous transposition of the spleen combined with ligation or constriction of the portal vein above its highest tributary.

TECHNIQUE

After induction with pentobarbital sodium and endotracheal intubation, a right sixth intercostal space thoracotomy was performed. The supradiaphragmatic inferior vena cava was freed within the mediastinum and was constricted to approximately 50 per cent of its circumference with either an aluminum band or heavy suture material encased in polyethylene tubing. The chest was closed in layers, and a catheter left in place to assure adequate inflation of the lung at the close of

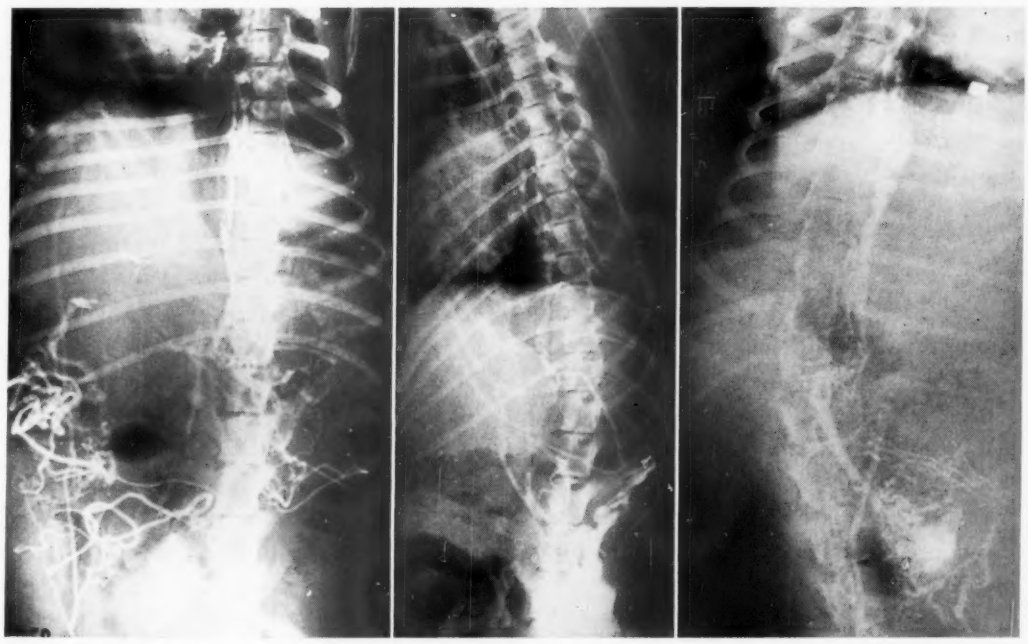

Fig. 1. Example of 3 splenoportograms performed at the conclusion of the final stage. Note the abnormal venous collateral vessels ascending beside the esophagus.

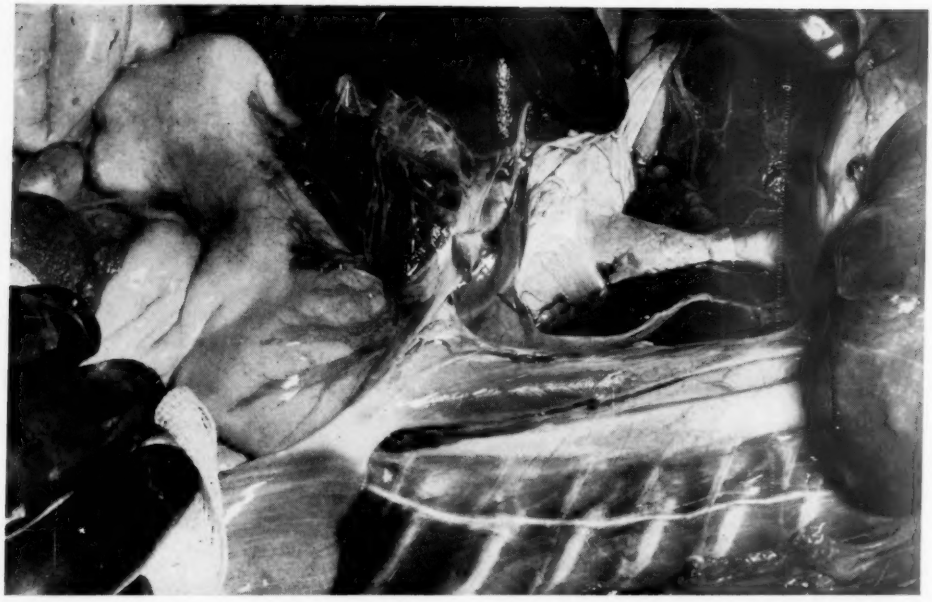

Fig. 2. Photograph taken at postmortem examination. Note aluminum band around the vena cava and abnormal venous collateral vessels lying in a periesophageal location.

the procedure. In 12 animals, this first stage procedure was followed after an interval of 2 to 4 weeks by repeat thoracotomy and the azygos vein was doubly ligated and interrupted below the superior intercostal vein. Of the 12 dogs, 5 survived. Since no apparent advantage was gained from completing the procedure in 3 stages, the last group of animals was treated by operation in 2 stages. Constriction of the supradiaphragmatic inferior vena cava was combined with azygos vein interruption in a single procedure. The second and final stage then consisted of subcutaneous transposition of the spleen and portal vein ligation or constriction.

The two-stage technique was employed in 18 animals, 11 of which survived. Half of these animals had omentectomy performed through a left paramedian incision at the time of the first stage operation to evaluate its possible effect on the development of collateral circulation.

Approximately 60 to 100 days after the completion of the first portion of the procedure, the 16 animals surviving the first procedures were reoperated upon. Fourteen of these had a one-stage ligation of the portal vein combined with subcutaneous transposition of the spleen. Prior to portal vein ligation, a 15 to 20 minute period of occlusion was allowed in order to correlate appearance of the bowel with future survival of the animal. In the other 2 animals, the bowel became cyanotic and went into spasm. It was believed that the animals could not survive complete occlusion and, in both cases, the portal vein was constricted by approximately 60 per cent with a heavy cotton ligature.

Of the 16 animals which underwent the final stage of the procedure, 6 animals survived. These were studied by splenoportography and esophagoscopy and were ultimately sacrificed and subjected to postmortem examination to establish the exact location of the abnormal venous collaterals which were found to develop.

These 6 animals included the 2 in which the portal vein was partially occluded. In 5 of the 6 animals splenoportography showed evidence of periesophageal varices. Esophagoscopy failed to demonstrate the presence of any submucosal varices. Autopsies performed at the conclusion of the experimental period demonstrated the presence of abnormal collateral veins lying in the adventitia of the esophagus. The 1 animal that did not develop varices was later found to

have had the portal vein ligated proximal to the junction with the splenic vein.

Ten animals failed to survive the final stage. Postmortem examination was performed on all of them and, in 1 case, splenoportography was performed immediately prior to death. Seven animals showed the same abnormal distribution of venous collateral shown by the animals which survived the second stage, but in the other 3 we were unable to find any evidence of such abnormal collaterals.

We were unable to demonstrate the presence of any submucosal varices resembling those in man or those recently reported in monkeys[2] which had undergone similar staged procedures.

SUMMARY

1. Forty dogs were subjected to a series of venous ligations designed to produce portal hypertension in the hope that they would develop collateral venous circulation in the form of esophageal varices.

2. Sixteen animals survived the first stage, which consisted of inferior vena cava constriction and azygos vein interruption.

3. Six animals survived the second stage which consisted of portal vein ligation or constriction.

4. Five of the 6 animals were shown by splenoportography to have abnormal venous collaterals running along the wall of the esophagus. Esophagoscopy failed to demonstrate the presence of any submucosal varices. One animal with ligation of the portal vein proximal to the junction with the splenic vein failed to develop such collaterals.

5. Of the 10 animals which failed to survive the final stage, 7 demonstrated periesophageal varices at autopsy, while 3 showed no evidence of these abnormal collaterals.

6. Omentectomy failed to add appreciably to the development of collateral circulation.

7. This experiment demonstrated the fact that, although dogs develop abnormal venous collaterals which lie along the outer wall of the esophagus in periesophageal tissue, the varices produced by this technique are not similar to those seen in human beings or monkeys with portal hypertension.

REFERENCES

1. Baronofsky, I. D., and Wangensteen, O. W.: Obstruction of splenic vein increases weight of stomach and predisposes to erosion or ulcer, Proc. Soc. Exper. Biol. & Med. **59:** 234, 1945.
2. Laufman, H., Bernhard, V., Roach, H., and Champlain, G.: Experimental production of esophageal varices in the *Macaca rhesus,* Surg. Gynec. & Obst. **110:** 4, 1960.
3. McKee, F. W., Schilling, J. A., Tischkoff, G. H., and Hyatt, R. E.: Experimental ascites, Surg. Gynec. & Obst. **89:** 1, 1949.
4. Milnes, R. F., and Child, C. G.: Acute occlusion by ligature of the portal vein in the *Macaca rhesus,* Proc. Soc. Exper. Biol. & Med. **70:** 332-34, 1949.
5. Rousselot, L. M., and Thompson, W. P. A.: A report on studies of the experimental production of congestive splenomegaly, Proc. Soc. Exper. Biol. & Med. **40:** 705, 1939.
6. Tamiya, T., and Thal, A. P.: Esophageal varices produced experimentally in the dog. Read before Forum on Fundamental Surgical Problems, American College of Surgeons, Atlantic City, Sept. 28, 1959.
7. Tamiya, T., and Thal, A. P.: Esophageal varices produced experimentally in the dog, Surg. Gynec. & Obst. **111:** 147-154, 1960.

Method for direct inspection of the right atrium: experimental investigation in the dog

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With current methods of examination it is often difficult to determine the precise location of certain anomalies involving the right atrium, such as septal defects, abnormal venous return, subendocardial fibroelastosis, and so on. A method which makes possible direct inspection of the atrium might therefore be of great help in the preoperative diagnosis. To enable such inspection to be made within the blood-filled heart we have devised an instrument which has been tried both in experiments with models and in experiments on animals. We have used for the purpose a 4 mm. right-angled telescope. Over the objective we slipped a thin rubber bladder which was inflatable through a long, thin catheter attached along the telescope. A thin metal shield on the back side of the rubber bladder prevented inflation of it elsewhere than over the objective (Fig. 1). To prevent damage in case of possible rupture of the bladder, the latter was filled with physiologic saline solution. If for optical reasons a gas is preferred, carbon dioxide may be used. The size of the rubber bladder may be varied, but even with inflation to a volume of 5 ml. its wall becomes completely transparent, as is apparent from Fig. 2, in which the objective can be clearly seen.

In the experiments carried out on models the instrument was introduced into a blood-filled glass cylinder. When the bladder was lightly pressed against the wall inside the vessel it was possible to read clearly the text on the outside of the glass, with a field of vision of about 1 cm.^{2} (Fig. 3).

In experiments on live dogs the instrument was introduced through the right external jugular vein and pushed down via the superior vena cava into the right atrium, after which the rubber bladder was inflated with about 5 ml. of physiologic saline solution. When the instrument was pressed

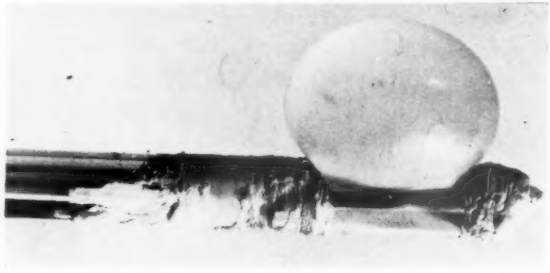

Fig. 1. Lateral picture of a right-angled telescope with a diameter of 4 mm. with an inflated rubber bladder over the objective.

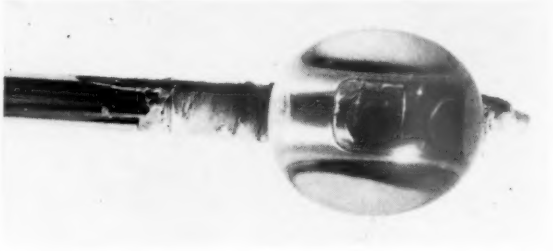

Fig. 2. The objective of the instrument may be seen through the completely transparent rubber bladder.

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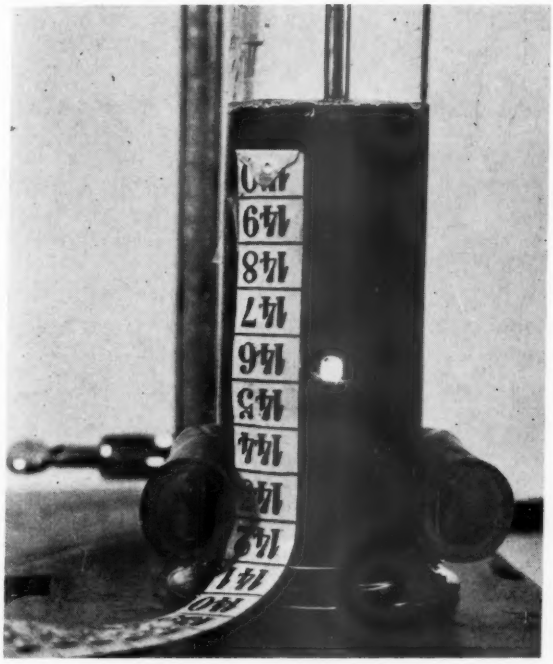


Fig. 3. The instrument was introduced into a blood-filled glass cylinder which was held in position by clamps. The rubber bladder was held pressed against the glass wall and displaced the blood.

lightly against the vascular or cardiac wall a good picture of the anatomic details was obtained. Thus it was easy to inspect the orifice of the azygos vein in the superior vena cava, the coronary sinus, trabeculae cordis in the auricle, the atrial septum, and the base and commissures to the tricuspid valves. In one experiment it was possible to get a clear view of an artificially produced septal perforation with left-to-right shunt. It was also possible for shorter periods to introduce the instrument into the right ventricle and see valves, papillary muscles, and chordae tendineae. The dogs tolerated the examination well and we had the instrument lying in the heart for inspection for several hours. The risk of air embolism was eliminated by a ligature tied around vein and instrument. The pulsations from the heart are transmitted through the instrument and any disturbances of rhythm were felt immediately. Extra systoles and transient attacks of tachycardia occurred during the passage through the valves and when the bladder

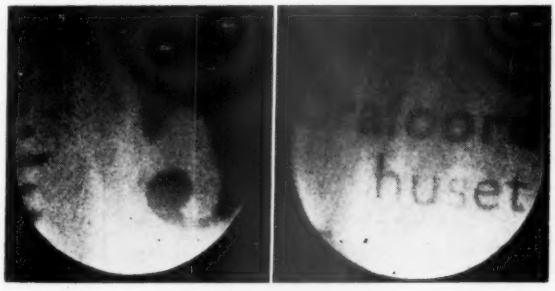
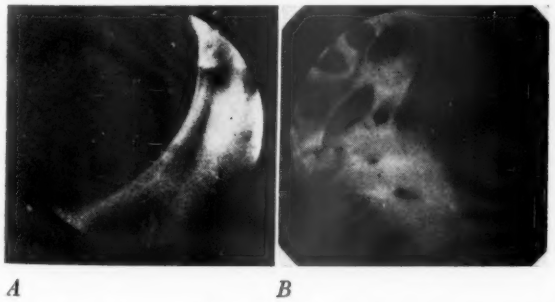
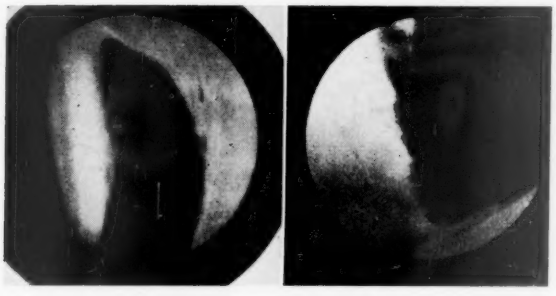


Fig. 4. Pictures which were obtained with the instrument in the blood-filled glass cylinder.



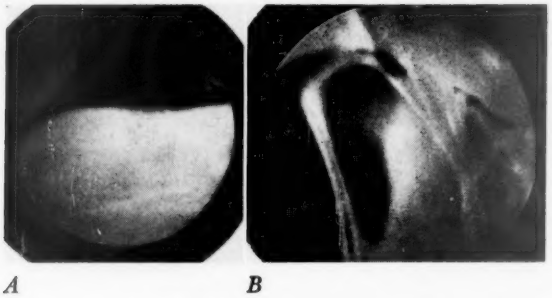
A *B*

Fig. 5. Photographs of blood-filled dog's heart at standstill. *A*, Aperture of azygos vein opening into superior vena cava. *B*, Trabeculae cordis in auricle.



A *B*

Fig. 6. *A*, Coronary sinus. *B*, Artificially produced defect in the atrial septum.



A *B*

Fig. 7. *A*, Tricuspid commissure. *B*, Papillary muscles and chordae tendineae from the right ventricle.

entirely occluded the inferior vena cava. It was not possible to observe any signs of stasis whatsoever from the upper half of the body. The brief disturbances of rhythm ceased as soon as the position of the instrument was altered. The instrument employed is completely satisfactory for purposes of inspection, but necessitates long exposure time for photography, as it is of too fine a caliber to hold flashlight equipment. To demonstrate the picture obtained on inspection, we have taken photographs of the blood-filled heart at a standstill. The black and white pictures give a good idea of the details of the living heart that are easily observable through the instrument. The exposure times were approximately 1 minute (Figs. 4 through 7).

SUMMARY

We have described a method for the inspection of the right atrium and an instrument constructed for this purpose. The method has been elaborated in experiments on models and on animals. The instrument consists of a telescope over the objective of which is fixed a rubber bladder inflatable with saline solution. The rubber bladder, which is completely transparent, displaces the blood and permits inspection of the cardiac or vascular wall.

Myocardial metabolism during perfusion of the coronary circulation with gaseous oxygen

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The maintenance of certain vertebrate tissues and organs in an active state by perfusion of the vascular system with gaseous oxygen has recently been demonstrated. In 1954 Bunzl and associates[1] showed that the reflex activity of the spinal cord of the frog could be maintained for periods of up to 24 hours when the arterial circulation was perfused with gaseous oxygen. Since then Burns and associates[2] have shown that the skeletal and cardiac muscle of the rabbit could be similarly maintained. Our interest in this subject originated with a group of experiments in which it was shown that the isolated dog heart continued to beat for periods of up to 8 hours when the coronary arteries were perfused solely with oxygen in the gaseous state.[11] The purpose of the present report is to describe experimental evidence of a continuation in the carbohydrate metabolism under these circumstances.

METHODS AND RESULTS

The isolated hearts of 34 dogs were used in the study and were divided into 3 experimental groups.

Group I (controls). Eight adult mongrel dogs were anesthetized with sodium pentobarbital and the left side of the chest was opened. The heart was rapidly removed, the atria were trimmed away, and the ventricles were weighed. The heart was quick-frozen in dry ice. Usually 3 minutes elapsed between the time of the incision and freezing; in each instance the time was less than 7 minutes. A sample of left ventricle (approximately 20 grams) was accurately weighed and homogenized for 10 minutes in 100 ml. of ice-cold 7 per cent perchloric acid in a Waring Blendor. The homogenate was separated by centrifugation for 30 minutes at 3,000 r.p.m. and the supernatant neutralized to pH 7 to 8 with 5 per cent potassium hydroxide. The potassium perchlorate was removed by centrifugation for 30 minutes at 3,000 r.p.m., and the resulting supernatant was used for glycogen and lactate determinations. Glycogen was precipitated twice from 95 per cent alcohol and determined by the anthrone colorimetric method used by Seifter and associates.[12] Lactate was determined enzymatically by the method of Horn and Bruns.[5] Duplicate determinations were made on each specimen.

The results of these determinations showed the control glycogen level to range from 260 to 705 mg. per 100 Gm. of left ventricle (wet weight) with an average of 533 mg. These figures are in agreement with previous ones reported in the literature.[7, 8, 14] The lactate values in these hearts ranged from 15 to 98 micromols per 100 Gm. of left ventricle, with an average of 58 micromols per 100 Gm.

Group II (isolated heart with oxygen perfusion of coronary circulation). The hearts of 11 dogs were removed rapidly as

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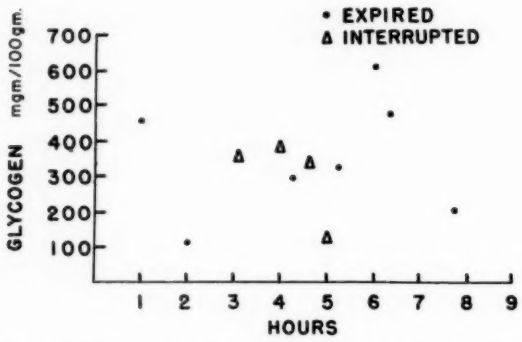

Fig. 1. Myocardial glycogen in hearts perfused with gaseous oxygen.

in the control group and cannulas were inserted into the right and left coronary arteries and sutured in place. The heart was placed in a humidified chamber of Plexiglas which was maintained constantly at 37° C. The coronary arteries were first perfused for a period of 1 minute with oxygenated donor blood from a reservoir and this was then flushed by the perfusion of 0.9 per cent saline solution. When the returns from the coronary sinus were clear (usually in approximately 1 minute) the perfusion was changed to gaseous oxygen (95 per cent) and carbon dioxide (5 per cent) from a cylinder. This technique has been described previously in detail.[11] Seven of the hearts were permitted to beat until spontaneous arrest or ventricular fibrillation occurred, and 4 others were interrupted at various periods. All hearts were quick-frozen and determinations performed as in the control hearts.

The results of glycogen determinations for isolated hearts perfused with gaseous oxygen for from 60 to 465 minutes are shown in Fig. 1. The values range from 114 to 610 mg. per 100 Gm. of ventricle, and 8 of the 11 showed a level in excess of 300 mg. per 100 Gm. The lactate concentrations are shown in Fig. 2 and demonstrate that the values are maintained at essentially normal levels, with a range of 4.4 to 76.0 and an average of 26 micromols per 100 Gm. of ventricle.

Group III (nonperfused hearts suspended at 37° C.) The hearts of 11 dogs were removed and cannulated in the same manner as in Group II. Nothing was perfused into the coronary circulation and the hearts were suspended in the Plexiglas chamber and permitted to arrest spontaneously and were observed for periods of 1 to 7 hours. The hearts were then quick-frozen and determinations performed.

The results showed that the myocardial glycogen level fell sharply after the first hour and after this time was usually less than 50 mg. per 100 Gm. of ventricle (Fig. 3). The values for the lactate determinations showed that in each instance the level was markedly elevated (164 to 297 micromols per 100 Gm. of ventricle). These data are shown in Fig. 2.

DISCUSSION

The utilization of oxygen by tissues and organs when perfused through the vascular system in the gaseous state is a basic physiologic problem and one which has been given recent study. In 1902 Magnus[6] first observed that the beat of the cat heart could in some instances be maintained for up to 1 hour by perfusion of gaseous oxygen. In 1954 Bunzl and associates[1] reported the observation that the spinal cord of the decerebrate frog could be maintained in a reflexly active state for periods of up to 24 hours when the vascular system was perfused with a mixture of oxygen (95 per cent) and carbon dioxide (5 per cent) in

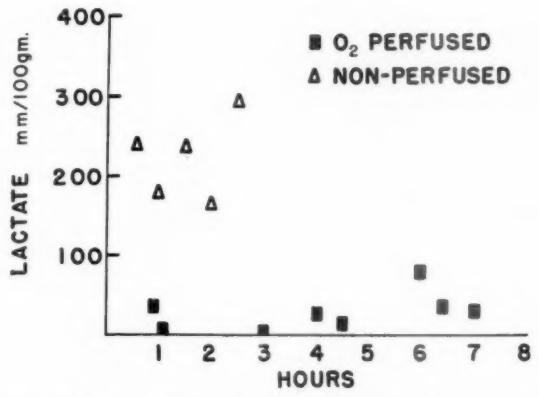

Fig. 2. Effect of gaseous oxygen perfusion on myocardial lactate.

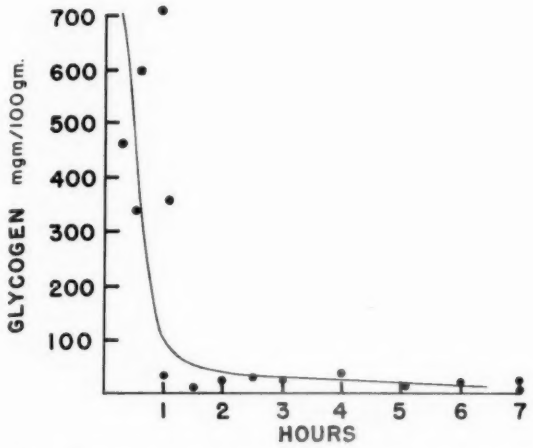

Fig. 3. Myocardial glycogen in nonperfused hearts suspended at 37° C.

the gaseous state. Additional studies on this problem by Burns, Robson, and Smith[2] showed that the isolated hind limb of the rabbit could be maintained in a physiologic state for 3 to 4 hours with oxygen perfusion of the femoral artery. These workers also reported that similar perfusion of the coronary circulation permitted the heartbeat to continue for a period of up to 3 hours. In our laboratory it has been shown that the isolated dog heart may continue to beat for periods of up to 8 hours with coronary arterial oxygen perfusion alone, and electrocardiographic activity may persist for a period of 12 hours.[11] An extension of these studies has shown that gaseous oxygen perfused retrograde into the coronary circulation through the coronary sinus also maintains the beat of the isolated dog heart, although less effectively than similar perfusion through the coronary arteries.[13] Although it appears clear that gaseous oxygen must be utilized by the cells under these circumstances, there has not yet been biochemical confirmation of this point.

The acid-soluble fraction of glycogen has been shown by several investigators to be the metabolically more active fraction in canine myocardium.[3, 4, 9, 10] This fraction has been measured under variable conditions of anoxia and shown to decrease. Merrick[7] demonstrated a decrease of approximately 45 per cent in 4 hours (396 to 210 mg. per cent) in areas of myocardium rendered anoxic by experimental coronary occlusion. Merrick and Meyer[8] reported a decrease from 565 mg. per cent to 128 mg. per cent in 2 hours in myocardium associated with hypoxia after induction of experimental pneumothorax. Tennant and associates[14] also reported a decrease in total myocardial glycogen after experimental coronary occlusion. In excised hearts, Merrick and Meyer[8] demonstrated a decrease in acid-soluble glycogen from 699 to 451 mg. per cent after 1 hour and to 207 mg. per cent after the hearts had been kept at 4° C. for 7½ hours. Conn, Wood, and Morales[4] demonstrated a decrease in myocardial glycogen from 475 mg. per cent to values approaching zero with a concomitant rise in lactate of approximately 400 per cent after 2 hours in hearts maintained at 37° C. These data indicate that under anaerobic conditions the myocardium breaks down glycogen to lactate and that, in this situation, glycogen stores are rapidly depleted.

In the hearts perfused with oxygen in the present study, the acid-soluble glycogen fraction remained above 200 mg. per cent after 7½ hours of continuous beating at 37° C. With the variation in glycogen stores known to occur in the normal dog heart, it is difficult to determine the rate of breakdown under the experimental conditions. These data indicate, however, that the beating isolated heart perfused with oxygen continues to utilize glycogen. The rapid depletion and low level of glycogen in nonperfused hearts suggest that anaerobic glycolysis is being carried out and that this becomes less effective in maintaining cardiac activity over an extended period of time.

The control values for lactate obtained in the present experiments are comparable to those reported by Tennant and associates and by Conn, Wood, and Morales. The lactate concentration in the nonperfused hearts maintained at 37° C. for several hours was considerably greater than the control values for the normal heart. The relatively slow depletion of glycogen and the absence of excessive accumulation of

lactate in hearts maintained with an active beat for periods of up to 7½ hours suggest that oxygen is utilized in cellular metabolism under these circumstances.

SUMMARY

The isolated dog heart continues to beat for prolonged periods when the coronary circulation is perfused solely with gaseous oxygen. Determinations of myocardial glycogen and lactate levels indicate that oxygen is metabolically utilized under these circumstances. In control preparations without oxygen perfusion, the glycogen is rapidly depleted with a marked increase in myocardial lactate. With oxygen perfusion, glycogen levels remained appreciable for prolonged periods and the lactate concentration persisted at essentially normal levels. These data indicate that oxygen is utilized by the myocardium in carbohydrate metabolism when perfused through the coronary circulation in the gaseous state.

REFERENCES

1. Bunzl, A., Burgen, A. S. V., Burns, B. D., Pedley, N., and Terroux, K. G.: Methods for studying the reflex activity of the frog's spinal cord, Brit. J. Pharmacol. **9:** 229, 1954.
2. Burns, B. D., Robson, J. G., and Smith, G. K.: The survival of mammalian tissues perfused with intravascular gas mixtures of oxygen and carbon dioxide, Canad. J. Biochem. Physiol. **36:** 499, 1958.
3. Carroll, N. V., Longley, R. W., and Roe, J. H.: The determination of glycogen in liver and muscle by use of anthrone reagent, J. Biol. Chem. **220:** 583, 1956.
4. Conn, H. L., Wood, J. C., and Morales, G. S.: Rate of change of myocardial glycogen and lactate following arrest of the coronary circulation, Circulation Res. **7:** 721, 1959.
5. Horn, H. D., and Bruns, F. H.: Quantitativ Bestimmung von L (+) - Michsaure mit Milchsauredehydrodgenase, Biochim. & biophys. acta **21:** 378, 1956.
6. Magnus, R.: Die Thatigkeit des Ueberlebenden Saugethierhergene bei Durchstromung mit Gasen, Arch. f. exper. Path. u. Pharmakol. **47:** 200, 1902.
7. Merrick, A. W.: Experimental coronary occlusion in dogs and its effect upon the cardiac glycogen fractions, Circulation Res. **5:** 435, 1957.
8. Merrick, A. W., and Meyer, D. K.: Glycogen fractions of cardiac muscle in the normal and anoxic heart, Am. J. Physiol. **177:** 441, 1954.
9. Meyer, D. K., Russell, P. L., Platner, W. S., Purdy, F. A., and Westfall, B. A.: Synthesis of glycogen fractions by heart homogenates, Proc. Soc. Exper. Biol. & Med. **90:** 15, 1955.
10. Russell, J. A., and Bloom, W. L.: Extractable and residual glycogen in tissues of the rat, Am. J. Physiol. **183:** 345, 1955.
11. Sabiston, D. C., Talbert, J. L., Riley, L. H., and Blalock, A.: Maintenance of the heart beat by perfusion of the coronary circulation with gaseous oxygen, Ann. Surg. **150:** 361, 1959.
12. Seifter, S., Dayton, S., Novic, B., and Muntwyler, E.: The estimation of glycogen with the anthrone method, Arch. Biochem. **25:** 191, 1950.
13. Talbert, J. L., Riley, L. H., Sabiston, D. C., and Blalock, A.: Retrograde perfusion of the coronary sinus with gaseous oxygen, Am. Surgeon **26:** 189, 1960.
14. Tennant, R., Grayzel, D. M., Sutherland, F. A., and Stringer, S. W.: Studies on experimental coronary occlusion. Chemical and anatomical changes in the myocardium after coronary ligation, Am. Heart J. **12:** 168, 1936.

Survival and physiologic studies with profound hypothermia (0° to 8°C.) and a pump-oxygenator in dogs

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The use of combined hypothermia and extracorporeal circulation for open-heart surgery has increased markedly in recent years. The experimental work of Gollan and his group[4-6] demonstrated the feasibility of this approach. Sealy[8, 9] and Brown[2] and their associates have reported a large series of open-heart operations in which this method was used. The control of ventricular fibrillation, the initial barrier, has been successfully achieved.[10] The obvious extension of the method is increasing dependence upon hypothermia, with simultaneous decreasing dependence upon the extracorporeal heart-lung apparatus, except in the induction and maintenance of hypothermia and restoration of normothermia. In general, the more profound the hypothermia the more time is afforded the operator in clinical situations for interruption of the circulation. Exploration of the factors involved in degrees of hypothermia which approach the end point, the freezing point of the body fluids, assumes significance.

Woodhall and colleagues[12] reported the case of a patient in whom body temperature was reduced to 4° C. with cessation of the entire circulation for 10 minutes. Rewarming was followed by complete recovery. Reports of hypothermia in the range of 9° to 20° C., with extracorporeal circulation and heat exchangers, are now appearing regularly. As has often happened in this fast-moving field, the clinical application of the technique has outrun the experimental and physiologic observations. A considerable body of data on the electrolyte changes, pH, oxygen concentration, and fluid shifts which occur during hypothermia has accumulated, but most of the studies have been performed at temperatures above 20° C.

Our study was an attempt to evaluate the effects of profound hypothermia between 0° and 8° C. on the physiologic processes and the survival rates in dogs. With a disk oxygenator, De Bakey pumps and a Brown-Harrison heat exchanger, cooling and warming were carried out by partial perfusion. The technique of cooling, utilizing partial perfusion, the survival rates, and studies of the blood electrolytes, blood pH, blood osmolarity, hematocrit, white blood cell count, and platelets are reported.

TECHNIQUE

Fifteen mongrel dogs which weighed between 15 and 26 kilograms were used in these studies. They were anesthetized intravenously with 25 mg. per kilogram of pentobarbital sodium and, after the insertion of an endotracheal tube, were attached to a positive-pressure respirator. The animals were then given heparin intravenously, 2

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mg. per kilogram of body weight, and quinidine intravenously, 20 mg. per kilogram of body weight. An equivalent amount of quinidine was added to the priming blood of the extracorporeal unit. Bardic plastic venous catheters, ranging in size from 18 to 24 Fr. were then inserted into the external jugular veins bilaterally, as well as into one femoral vein early in the study and into both femoral veins later in the study. These catheters were passed as far as possible into the superior and inferior venae cavae. A catheter for blood pressure recordings was inserted into one of the femoral arteries and an inflow cannula for the return of blood from the heart-lung machine was placed in the other femoral artery. Electrocardiogram leads were attached and a temperature probe was placed into the dog's rectum. In some instances temperature probes were inserted into the esophagus, left side of the heart, and right side of the heart.

A 12 inch disk oxygenator with De Bakey pumps and a single Brown-Harrison heat exchanger was used. This apparatus was cleaned with saturated sodium hydroxide solution prior to each run and cold sterilization carried out with ethylene oxide gas. The Brown-Harrison heat exchanger was connected to water taps and to an ice unit for the circulation of ice water or brine solution. Mayon tubing was used and connectors were stainless steel or Teflon.

Partial perfusion was begun with the blood flow regulated according to the venous gravity return. The return reservoir was approximately 15 to 20 inches below the level of the operating table. Oxygen flow varied from 5 to 8 L. per minute and the disks rotated at 80 to 120 r.p.m. From the ice bucket, water or brine ranging from 3° to −10° C., was circulated through the heat exchanger. Ice chips were then placed over the body of the dog in order to reduce the absorption of heat by the body surface of the cooled animal. The dogs were cooled as rapidly as possible to rectal temperatures which varied between 0° and 8° C. The extracorporeal unit continued to circulate blood through the animal during the entire period of hypothermia. The respirator was slowed or stopped when the dog's heart action ceased. After 10 to 30 minutes at the lowest temperature achieved, the animals were rewarmed with tap water of about 42° C. The respirator was started at the first sign of cardiac activity. When the heartbeats were effective and rectal temperatures had returned to 34° C., partial perfusion was discontinued. Supportive therapy, in the form of sodium bicarbonate and norepinephrine administration and continuous positive respirations, was instituted when necessary. In 3 of the animals, ventricular fibrillation occurred. In these dogs the chest was opened and cardiac massage and internal defibrillation carried out. When the animals appeared to be responding satisfactorily, decannulation was completed and the animals were observed for a period of 2 weeks.

Before, during, and after the bypass, samples of blood were taken for plasma electrolyte, plasma hemoglobin, blood pH, total protein, blood osmolarity, platelet, and white cell determinations. These samples were drawn at normothermia, 30°, 20°, 10° and 5° C., and at the lowest temperature reached during cooling. During rewarming, samples were obtained at 5°, 10°, 20° and 30° C., and hourly during the recovery of the animal.

RESULTS

Fifteen dogs were cooled by means of the Brown-Harrison heat exchanger and the extracorporeal apparatus described. Eight of the 15 animals were cooled to rectal temperatures between 3° and 8° C. for periods of time ranging from 10 to 30 minutes. In this group there were 6 dogs that survived for 2 weeks or more and were then sacrificed. These were considered complete survivors. Seven animals were cooled to between 0° and 3° C. with no long-term survivors (Table I).

In the dogs cooled to between 3° and 8° C., the rectal temperatures of the 6 complete survivors were 3°, 3°, 4°, 5°, 7°, and 8° C. The dogs were maintained at the lowest temperature reached for periods of time

varying from 10 to 30 minutes. One of these complete survivors was kept at 5° C. for 30 minutes. In Dog. No. 41 the rectal temperature was maintained at 3° C. for 30 minutes and was at 5° C. or below for 1 hour with complete survival. The longest period of partial perfusion, which includes cooling, maintenance at the low temperature, and rewarming, was approximately 3 hours and 25 minutes. The period of cardiac arrest in the dogs that survived indefinitely ranged from 20 minutes to 2 hours and 20 minutes, with a mean of 79 minutes.

The last electrical heartbeats in all dogs were recorded between 7° and 16° C. In Dog No. 24 (Table I), which survived indefinitely, fibrillation occurred during the cooling period at 27° C., but during the rewarming phase normal sinus rhythm returned with the onset of electrical activity. This animal was kept at 4° C. for 10 minutes and underwent partial perfusion for 1 hour and 40 minutes. The hearts in none of the animals which survived indefinitely fibrillated during rewarming. The first heartbeat recorded on the electrocardiogram occurred at temperatures which ranged from 10° to 20° C. This first electrocardiographic impulse did not correlate with the start of effective myocardial contraction. There was usually a 5° to 10° C. temperature difference between the beginning of the first heartbeat registered electrocardiographically and the first evidence of satisfactory myocardial contraction as indicated by a recordable blood pressure.

Two animals cooled to 3° and 3.5° C. survived 36 and 32 hours, respectively. The hearts of these animals had not fibrillated and the time of cardiac arrest was 75 minutes in both cases. Both animals had fully reacted prior to their deaths.

The average rectal temperature was 21° C. at the end of the first 10 minutes of cooling and 12° C. at the end of the first 20 minutes of perfusion. Longer periods of time were necessary to obtain lower temperatures and up to 1 hour was required in most cases to reach temperature ranges below 5° C. This was especially true in cases in which ice water without salt was used for the refrigerant. On the other hand, rewarming was more rapid, and in most cases 30° C. could be reached in approximately 30 minutes.

There were 7 attempts made to lower the rectal temperature below 3° C. The longest survival in this group was 24 hours in a dog which had been cooled to 1° C. for 10 minutes. The other 6 animals in this series

Table I. *Data on 15 dogs (hypothermia from 0° to 8° C.)*

Dog No.	Lowest rectal temperature (C.)	Total time at		Rectal temperature at		Period of		
		Lowest temperature (min.)	5° C. or below (min.)	Last electrical activity of heart (C.)	First electrical activity of heart (C.)	Cardiac arrest (min.)	Perfusion (min.)	Survival (hr.)
20	7	10	0	14	20	20	40	Complete
22	8	10	0		19	30	80	Complete
24	4	10	13	(27*) 12	15	65	100	Complete
25	1	10	37	(21*) 10	11*	90	180	10
26	1	12	43	13	13*	80	150	24
27	3	20	35	16	11	80	165	36
28	1	10	45	13	9.5	70	130	4
32	3	10	10	10	15	30	55	Complete
33	0	4	30	14	19	85	140	4
34	2	10	33	15	15	60	120	3.5
36	3.5	30	40	16	8	80	148	32
37	1	12	35	16	9	62	110	5
39	5	30	30	7	11	50	120	Complete
41	3	30	70	14	10	140	205	Complete
42	2	25	120	16	19*	150	240	5

*Ventricular fibrillation.

Table II. *Data on Dog No. 41 (hypothermia to 3° C., complete survival)**

Rectal temperature (C.)	*Time (min.)*	*Electrolytes*				*Hematocrit*	*pH*	*Osmolarity (milliosmoles)*	*WBC*	*Platelets*
		Na+	*K+*	*CO_2*	*Cl−*					
36.6	0	145.2	3.0	25.2	114	43	7.41	312	7600	149,000
20	15	145.9	3.5	23.2	110.5	39	7.35	324	—	—
10	40	145.9	3.8	27.0	107.7	39	7.32	324	—	—
5	80	144.4	3.89	29.2	107.0	40	7.30	327	—	—
3.5	115	143.4	4.1	29.4	104.7	41	7.27	332	—	—
3	160	143.2	4.4	30.2	103.6	42	7.24	338	1900	12,000
20	180	143.9	4.1	27.0	105.7	43	7.26	332	—	—
25	187	145.4	4.0	25.2	106.9	44	7.26	326	—	—
30	195	146.4	3.9	23.8	107.7	44	7.27	324	5500	42,000
35†	320	147.1	3.4	20.4	116.8	53	7.3	318	9400	76,000

*Ice water only in heat exchanger.

†Two hours after end of perfusion.

were cooled to 0°, 1°, 1°, 1°, 2°, and 2° C. with survival periods of 4, 10, 4, 5, 3½, and 5 hours, respectively. Ventricular fibrillation during rewarming occurred in 3 of the animals cooled below 3° C., but the hearts of all were easily defibrillated. The average time of partial perfusion in animals cooled below 3° C. was somewhat longer than in the first group. In all but one of the attempts to cool below 3° C., salt was added to the circulating ice water in an effort to produce a greater temperature differential between the blood in the heat exchanger and the solution being used for cooling. These efforts resulted in the circulation of brine at temperatures of −5° to −10° C. In several of these experiments the blood flow through the heat exchanger appeared to slow, and in one experiment there appeared to be complete obstruction in the heat exchanger. This was true in Run No. 27, at which time the brine in the ice bucket was at a temperature lower than −10° C. It was therefore necessary to circulate through the heat exchanger a warming solution before adequate blood flow would again pass through it. One animal in which the brine solution was recorded as −10° C. had a rectal temperature of 0° C. for approximately 10 minutes. There was recovery of the heart and a survival for 4 hours, although it was necessary to use Adrenalin and Levophed during the rewarming phase. It should be noted that in 4 of the 6 dogs which survived indefinitely, brine between −3° and −6° C. was also used in the heat exchanger. The attempts to cool dogs below a rectal temperature of 3° C. were unsuccessful in terms of complete survival.

Serum sodium potassium, carbon dioxide, and chloride determinations were made during the period of cooling and rewarming. In all of the animals there was no doubt that during the period of cooling the serum potassium level rose progressively on an average of 1 to 1.4 mEq. per liter. During rewarming the level returned toward control levels. No significant change in serum carbon dioxide and sodium levels was found. The level of serum chloride demonstrated a small but significant decrease during the period of cooling and returned to normal during rewarming. There was no difference in electrolyte determinations between animals that survived indefinitely and had rectal temperatures above 3° C. and animals that only survived for 24 hours or less and had rectal temperatures ranging between 0° and 3° C. (Tables II and III).

The pH recorded during all phases of cooling and rewarming showed a consistent fall during the period of cooling, reaching the lowest point at the most profound level of hypothermia (7.12 to 7.24), and returning toward normal during the period of rewarming. There was no correlation between

the fall in pH and survival. Two of the dogs that survived indefinitely had pH falls to 7.12, whereas several of the animals in which the temperatures were taken below 3° C. and survival ranged from 3½ to 24 hours, had pH's recorded no lower than 7.23. The pH always returned toward normal limits during the rewarming phase, although it never actually reached the precooling levels. The pH was determined at normal blood temperature (37° C.) with a Cambridge constant temperature pH meter.[7]

There was a constant elevation of serum osmolarity during hypothermia, averaging an increase of 20 to 30 milliosmoles as compared to the preperfusion levels. During the rewarming phase the osmolarity of the blood returned toward normal, and, in fact, 2 hours after the end of the perfusion the osmolarity was generally at preperfusion levels. Again, no distinction can be made between the animals with indefinite survival and those in which the survival was of short duration and the rectal temperatures were below 3° C.

Hematocrits recorded during the period of cooling and rewarming either remained constant or showed a gradual tendency to rise slightly during the rewarming period. White blood cell counts showed a drop in white blood cell levels during profound hypothermia and a return toward higher levels during rewarming. Platelets dropped markedly at low temperatures to ranges of 400 to 12,000 and then returned toward normal limits during the warming phase, but did not approach the 100,000 range until 1 to 2 hours after the termination of perfusion. Total proteins rose minimally during hypothermia and remained elevated slightly during warming.

Plasma hemoglobin values were determined in 7 dogs. When the brine in the heat exchanger was maintained below –5° C. there was an increased hemolysis of blood. However, when the temperature of the brine was maintained at levels above –5° C., plasma hemoglobin levels were not excessive. The greatest amount of hemolysis recorded, in a dog in which the temperature of the heat exchanger was between 0° and –2° C., was 205 mg. per cent after 240 minutes of partial perfusion.

Blood flow was usually betwen 60 and 80 ml. per kilogram of body weight per minute at the start of partial perfusion and was regulated to match the gravity return flow. As cooling progressed, flow was continually decreased because of decreasing return so that below 5° C. blood flow was usually 25 to 45 ml. per kilogram of body weight. Any attempt to increase the flow would require the addition of large quantities of blood. The blood pressure decreased as flow de-

Table III. *Data on Dog No. 37 (hypothermia to 1° C., 5-hour survival)**

Rectal temperature (C.)	*Time (min.)*	*Electrolytes*				*Hematocrit*	*pH*	*Osmolarity (milliosmoles)*	*WBC*	*Platelets*
		Na +	*K +*	*CO_2*	*Cl –*					
35	0	147.6	3.3	21.4	118.2	40	7.46	310	—	—
20	15	146.9	4.0	20.8	115.8	35	7.33	326	1700	49,400
10	25	145.6	4.3	22.6	115.0	35	7.31	333	1400	10,000
5	40	143.7	4.7	24.4	114	37	7.30	342	—	—
2	54	143.2	5.5	24.4	111.3	37	7.24	346	—	—
1	60	142.7	5.9	24.6	105.7	36	7.21	350	1600	2,800
1	70	139.6	5.9	24.8	102.6	38	7.20	353	—	—
5	75	140.3	5.9	22.6	111.6	36	7.22	340	—	—
10	80	142.2	5.3	21.4	115.0	38	7.24	323	1400	4,800
20	89	144.9	4.9	20.2	125.1	40	7.27	323	—	—
30	100	145.6	4.2	17.8	130.4	41	7.30	323	1500	52,000
34†	230	148.1	3.3	12.4	124.0	56	7.33	310	—	—

*Brine to –5° C. used in heat exchanger.

†Two hours after end of perfusion.

creased and the heartbeat became ineffectual. At the lowest temperature ranges, the blood pressure maintained by the pump was recorded between 40 and 80 mm. Hg. During the rewarming phase, both flow and blood pressure gradually rose and, when the heartbeat returned and became effective, the blood pressure usually returned to prehypothermia levels.

DISCUSSION

The most intriguing result in these experiments was that none of the 7 animals in which body temperature was reduced below 3° C. survived for periods longer than 24 hours. On the other hand, of the 8 animals in which body temperatures were kept between 3° and 8° C., indefinite survivals were obtained in 6. The survival time in the 2 animals that died was 36 and 32 hours with body temperatures reduced to 3° and 3.5° C., respectively. We have been unable to determine any differences in electrolyte shifts or hemolysis between the animals which survived and the ones which died.

It has been repeatedly demonstrated that rats, mice, and hamsters[1, 11] can have body temperatures reduced to 0° C., or slightly below, for periods of up to 30 minutes with subsequent resuscitation and long-term survivals. It is our present belief despite our results to date, that there is no mystical significance about the level of 3° C. However, as attempts were made to reduce the body temperatures to this extreme range, technical factors were multiplied. As the effort to obtain lower temperatures was made, the differential temperature between the animal and the heat exchanger became progressively smaller and the period of time on the pump became longer. Decreasing return flow from the dog and gain of heat from the atmosphere became problems at extremely low temperatures.

One method whereby the rate of cooling can be increased is to decrease the temperature in the heat exchanger to below 0° C. As we have indicated, this was done in 6 of the 7 animals cooled to under 3° C. It was readily demonstrated that if the temperature of the heat exchanger was maintained at less than –5° C., marked hemolysis of blood occurred. In the range of 0° and –5° C. we found no significant increase in hemolysis. Nevertheless, there may have been other damage inflicted upon the blood at these low temperatures which we failed to determine. The technical limit of the reduction of temperature in the heat exchanger was –10° to –11° C. At this temperature, blood in the exchanger formed a sludge which would not circulate. With the exception of the 2 experiments in which very low temperatures were maintained in the heat exchanger, the degree of hemolysis was surprisingly small for the periods of time that the animals were on the pump. Indeed, it was somewhat less than one would expect under conditions of normothermia. This was probably due to the decreased volume of blood pumped by the extracorporeal unit during the lower levels of hypothermia.

During hypothermia below 10° C., return flows from the dog decreased markedly to ranges of 400 to 800 ml. and continued to decrease as extremely low levels of temperatures were approached. The cause of this apparent decrease in circulating pumped blood with profound hypothermia requires additional investigation. It is quite possible that sequestration of the blood in the animals' capillaries caused the decreasing return flow, but no definite evidence of such sequestration was sought. When the animal was returned to higher temperatures, flow increased considerably and, presumably, sequestrated blood and plasma were returned into the general circulation.

The only definite electrolyte changes noted were a rise in potassium level and a fall in chloride level. In vitro experiments on tissue sections have demonstrated that a lowering of temperature of isolated tissues usually results in a shift of potassium out of the cell and sodium into the cell.[3] It is possible that if hypothermia had been maintained for longer periods more dramatic electrolyte shifts would have been noted. Extracellular shifts in potassium are often more easily detected than shifts of sodium

since the normal amount of potassium in the circulating plasma is so small that relatively small additions are readily apparent. On the other hand, shifts in sodium are less easily noted, both because sodium is present in much greater concentration and because bone sodium may be mobilized rapidly to replace losses in extracellular sodium. Therefore, it is probably true that the changes found reflect a decreased activity of the cellular membrane at these low temperatures, with early shifts of potassium out of the cell, despite the fact that all components of these changes were not detected.

The rise in osmolarity found to occur in the course of these experiments, with a return to normal as the animal's temperature was brought to normal remains unexplained. Presumably, this could reflect a dissociation of various molecules at these low temperatures which would contribute additional osmoles to the plasma. The added osmoles were not reflected in any of the electrolytes measured in this study and further determination of nonprotein nitrogen and other plasma constituents will be required.

SUMMARY

Fifteen dogs were cooled to temperatures between 0° and 8° C. with partial perfusion and a heat exchanger. In animals cooled to between 3° and 8° C., survival was achieved in 6 out of 8. Dogs cooled below 3° C. failed to survive beyond 36 hours. Studies of serum electrolytes, blood osmolarity, hematocrit, and platelet count failed to reveal any major difference between animals which survived and those which did not.

REFERENCES

1. Andjus, R. K., and Smith, A. U.: Reanimation of adult rats from body temperatures between 0° and +2° C., J. Physiol. **128:** 446, 1955.
2. Brown, I. W., Jr., Smith, W. W., Young, W. G., Jr., and Sealy, W. C.: Experimental and clinical studies of controlled hypothermia rapidly produced and corrected by a blood heat exchanger during extracorporeal circulation, J. Thoracic Surg. **36:** 497, 1958.
3. Conway, E. J.: Nature and significance of concentrations relations of potassium and sodium ions in skeletal muscle, Physiol. Rev. **37:** 84, 1957.
4. Gollan, F., Blos, P., and Shuman, H.: Exclusion of the heart and lungs from circulation of hypothermic closed chest dogs by means of a pump oxygenator, J. Appl. Physiol. **5:** 180, 1952.
5. Gollan, F., Blos, P., and Shuman, H.: Studies in hypothermia by means of a pump oxygenator, Ann. Physiol. **171:** 331, 1952.
6. Gollan, F., Hamilton, E. C., Meneely, G. R.: Consecutive survival of open-chest, hypothermic dogs after prolonged bypass of heart and lungs by means of a pump-oxygenator, SURGERY **35:** 88, 1954.
7. Rosenthal, T. B.: The effect of temperature on the pH of blood and plasma in vitro, J. Biol. Chem. **173:** 25, 1948.
8. Sealy, W. C., Brown, I. W., Jr., and Young, W. G., Jr.: Report on the use of both extracorporeal circulation and hypothermia for open heart surgery, Ann. Surg. **147:** 603, 1958.
9. Sealy, W. C., Brown, I. W., Jr., Young, W. G., Jr., Smith, W. W., and Lesage, A. M.: Hypothermia and extracorporeal circulation for open heart surgery, Ann. Surg. **150:** 627, 1959.
10. Sealy, W. C., Johnson, P. A., Lesage, A. M., and Young, W. G., Jr.: Effect of quinidine on ventricular fibrillation in hypothermic dogs, Proc. Soc. Exper. Biol. & Med. **101:** 456, 1959.
11. Smith, A. U., Lovelock, J. E., and Parkes, A. S.: Resuscitation of hamsters after supercooling or partial crystallization of body temperatures below 0° C., Nature, London **173:** 1136, 1954.
12. Woodhall, B., Sealy, W. C., Kenneth, H. D., and Floyd, W. L.: Craniotomy under conditions of quinidine protected cardioplegia and profound hypothermia, Ann. Surg. **152:** 37, 1960.

The effect of thyroidectomy on experimental ascites

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Extensive clinical and experimental investigation has been undertaken in order to elucidate further the humoral factors involved in the production of ascites. In recent years there has been particular interest in the increased level of urinary aldosterone in patients with cirrhotic ascites[10] and in animals with experimental ascites.[5] It has been shown that adrenalectomy[6] or hypophysectomy[3] ameliorates ascites in experimental animals. Similarly, these procedures, when performed prophylactically, appear to protect the animals against ascites formation.[4, 15] In our laboratory interest has been primarily focused on the relation of the thyroid gland to the development of experimental ascites and its role in the thyroid-adrenal axis. We have demonstrated the amelioration of experimental ascites by surgical thyroidectomy.[1, 9] In the previous reports, ascites was produced by supradiaphragmatic constriction of the inferior vena cava. The present study is a further investigation of the effect of thyroidectomy on the course of experimental ascites produced by this method, and an investigation of the response to thyroidectomy of dogs with ascites secondary to tricuspid insufficiency and pulmonary stenosis. In addition, the influence of preliminary surgical or radiation thyroidectomy upon the subsequent production of ascites by supradiaphragmatic caval constriction is evaluated.

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MATERIALS AND METHODS

Four groups of experiments were performed in male and female mongrel dogs which weighed from 12 to 20 kilograms. Throughout the study, the animals were kept in individual metabolism cages. Food rations provided 30 mEq. of sodium daily and water ad libitum to 2,500 ml. Daily weight and urine volumes were measured and the urinary excretion of sodium was determined daily by flame photometry. Ascites formation was quantitated by abdominal palpation and measurements of weight gain and abdominal girth. All dogs were evaluated in this manner for a minimum of 10 days prior to each experimental study.

In the first group of 9 dogs, ascites formation was induced by partial constriction of the supradiaphragmatic portion of the inferior vena cava.[11] When the ascitic state was well established, 4 to 6 weeks after caval constriction, total surgical thyroidectomy with reimplantation of the parathyroid glands was performed. Metabolic and clinical observations were continued for from 1 to 3 months after thyroidectomy.

In the second group of 3 dogs, ascites was produced by combined tricuspid insufficiency

Table I. *Effect of thyroidectomy on experimental ascites produced by caval constriction or tricuspid insufficiency and pulmonary stenosis*

No. of dogs	*Operation*	*Clinical results*
5	Supradiaphragmatic inferior vena caval constriction followed by thyroidectomy	No detectable ascites or ascites significantly improved
4	Supradiaphragmatic inferior vena caval constriction followed by thyroidectomy	No significant change in amount of ascites
2	Tricuspid insufficiency and pulmonary stenosis followed by thyroidectomy	No detectable ascites
1	Tricuspid insufficiency and pulmonary stenosis followed by thyroidectomy	No significant change in amount of ascites

and pulmonary stenosis. Tricuspid avulsion was performed by the method of Sprafka and associates.[14] In approximately 2 weeks, when the dogs had recovered from the operation, pulmonary stenosis was produced by the technique described by Veith and Thrower.[16] Total thyroidectomy was performed after the animals had developed severe ascites. After thyroidectomy, the usual metabolic and clinical observations were resumed for at least 6 weeks.

In the third group of 8 dogs, thyroidectomy was performed from 3 days to 6 months prior to caval constriction. The animals were observed for 6 weeks after caval constriction.

In the fourth group of 4 dogs, preliminary thyroidectomy was accomplished by the administration of radioactive iodine (I^{131}). Two to 6 weeks prior to caval constriction, 24 to 40 mc. of I^{131} was administered intramuscularly as a single dose. The retained dose was approximately 10 to 15 mc., or 30,000 to 45,000 rads to the thyroid gland. The status of thyroid function was evaluated by serial radioactive iodine uptake determinations. A tracer dose of 25 to 40 μc of I^{131} was administered intramuscularly, and counted 48 hours later by a bismuth-walled Geiger-Mueller tube 21 cm. from the dog's neck. Metabolic observations were continued for 8 weeks after caval constriction.

RESULTS

Group I. All 9 dogs that had been subjected to caval constriction developed ascites within 3 to 4 weeks. This period was marked by a striking positive sodium balance with a gradual increase in body weight and abdominal girth. After surgical thyroidectomy, a significant loss of body weight and abdominal girth occurred in 5 dogs in from 3 to 10 weeks. These changes were accompanied by a significant increase in sodium excretion. In the 4 remaining dogs, there was no significant change in the amount of ascites (Table I). The results obtained in a previous group of 20 dogs studied in this manner are presented in Table II.

Group II. Two of the 3 dogs in which ascites had been produced by tricuspid insufficiency and pulmonary stenosis responded to surgical thyroidectomy with complete remission of ascites. The clinical improvement was accompanied by a striking increase in urinary sodium and water excretion 1 to 2 weeks after thyroidectomy (Fig. 1). Ascites was no longer clinically detectable within 1 month. The third animal exhibited no significant changes in the ascitic state (Table I).

Group III. All 8 dogs subjected to caval constriction after surgical thyroidectomy developed ascites within 3 to 4 weeks. This progressed to a severe degree and was as-

Table II. *Effect of thyroidectomy on experimental ascites produced by caval constriction**

No. of dogs	*Clinical results of thyroidectomy*
13	No detectable ascites
3	Ascites significantly improved but minimally present
4	No significant change in amount of ascites

*Compiled from article by Kreel and associates.[9]

sociated with the typical pattern of sodium retention. Six dogs observed for 6 weeks after caval constriction showed no evidence of loss of ascites. Two animals which were observed for as long as 3 months after caval constriction also failed to exhibit any clinical sign of loss of ascites (Table III).

Group IV. In the 4 dogs that received I^{131}, confirmatory evidence of ablation of thyroid function was obtained by means of I^{131} uptake studies. The caval constriction was performed 3 weeks after the therapeutic dose of I^{131} had been given. Microscopic examination at postmortem showed fibrosis of the thyroid gland. However, in all 4 animals the typical metabolic and clinical pattern of ascites ensued after caval constriction (Table III).

DISCUSSION

The results tabulated indicate that a significant number of dogs with experimental ascites produced by caval constriction will undergo remission after surgical thyroidectomy. This is in agreement with the previous reports from this laboratory (Table II). In addition, this effect has also been demonstrated to occur when the ascites is produced by tricuspid insufficiency and pulmonary stenosis. The latter preparation was used because spontaneous remissions of ascites produced by caval constriction have been observed.[8] Spontaneous remission of ascites produced by tricuspid insufficiency and pulmonary stenosis has rarely been reported.[17] Craig and colleagues[2] have recently noted remissions of experimental ascites after radiation thyroidectomy in dogs with ascites produced by tricuspid insufficiency and pulmonary stenosis.

All the animals in Groups III and IV which had been subjected to either radiation or surgical thyroidectomy prior to caval constriction developed ascites. This is in contrast to the beneficial effect of preliminary hypophysectomy or adrenalectomy.

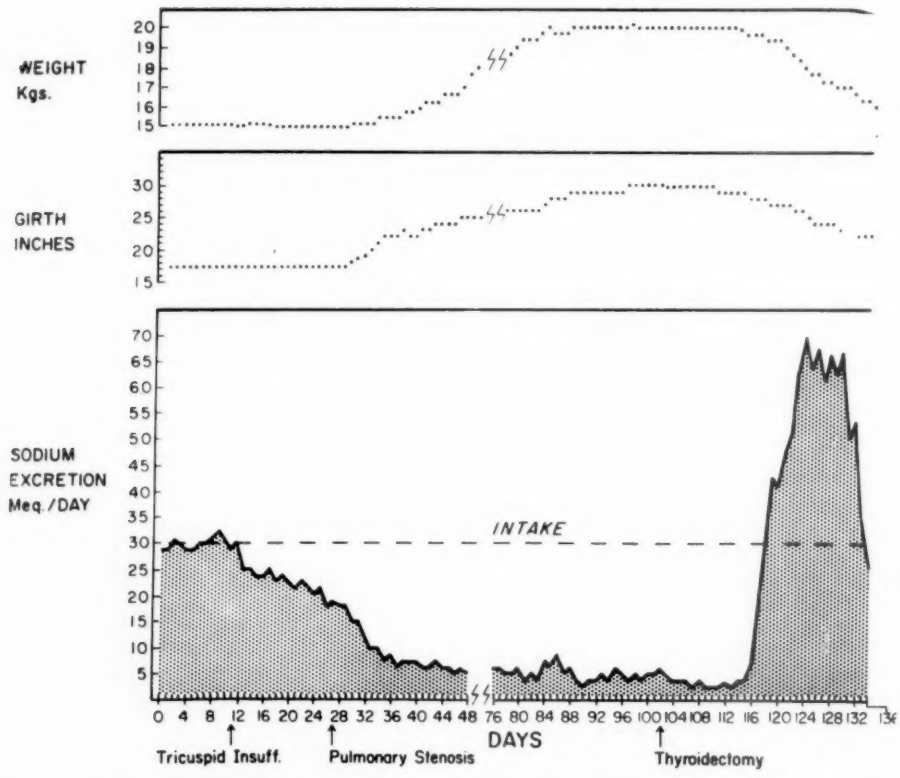

Fig. 1. Daily urinary sodium excretion, weight, and abdominal girth in an animal with ascites produced by tricuspid insufficiency and pulmonary stenosis which was completely relieved by thyroidectomy.

Table III. *Effect of preliminary surgical or radiation thyroidectomy on the production of ascites by supradiaphragmatic constriction of the inferior vena cava*

No. of dogs	*Operation*	*Clinical results*
4	Radiation thyroidectomy prior to supradiaphragmatic inferior vena caval constriction	Development of ascites
8	Surgical thyroidectomy prior to supradiaphragmatic inferior vena caval constriction	Development of ascites

The mechanism of these results is not entirely clear. However, we believe that a tenable hypothesis can be proposed. Animal studies have shown that there is a direct relationship between adrenal size and thyroid activity.[12] Peterson[13] has demonstrated the rate of synthesis of cortisol to be increased in thyrotoxicosis and decreased in myxedema. We may speculate that, similarly, the level of aldosterone may vary directly with thyroid activity. This would be consistent with the findings of Canter and his group,[1] who noted reduction in urinary aldosterone levels after thyroidectomy in ascitic dogs which underwent remission. These findings suggest that in the presence of secondary hyperaldosteronism, the adrenal gland becomes more sensitive to variations in thyroid function. Thus, ablation of thyroid function in these instances would produce a significant reduction in the elaboration of aldosterone.

The efficacy of preliminary adrenalectomy is evident because the source of aldosterone secretion is thus removed. Similarly, in the light of Davis'[7] recent report suggesting the ACTH dependence of aldosterone secretion, it is apparent that preliminary hypophysectomy could prevent the elaboration of ACTH and, secondarily, of aldosterone, thereby inhibiting ascites formation. Although it is probable that thyroidectomy results in some hypofunction of the normal adrenal cortex, the latter apparently remains able to respond to caval constriction with increased elaboration of aldosterone.

SUMMARY AND CONCLUSIONS

1. Amelioration of experimental ascites produced by supradiaphragmatic constriction of the inferior cava occurred in 5 of 9 dogs after total thyroidectomy.

2. Thyroidectomy caused the remission of ascites produced by tricuspid insufficiency and pulmonary stenosis in 2 of 3 dogs. This was accompanied by an increase in urinary sodium and water excretion.

3. Ascites formation was not affected by surgical or radiation thyroidectomy performed prior to caval constriction.

4. A mechanism which may be the cause of these results is presented.

REFERENCES

1. Canter, J. W., Kreel, I., Segal, R. L., Frankel, A., and Baronofsky, I. D.: Influence of thyroidectomy on experimental ascites, Proc. Soc. Exper. Biol. & Med. **100:** 771, 1959.
2. Craig, R. L., Ranney, R. E., Wetzel, N., and Bianchi, R. G.: The effect of radiothyroidectomy upon experimental ascites, Clin. Res. **8:** 237, 1960.
3. Craig, R. L., and Wetzel, N.: The effect of hypophysectomy on ascites due to circulatory failure in the dog, Quart. Bull. Northwestern Univ. M. School **28:** 55, 1954.
4. Craig, R. L., Wetzel, N., and Bianchi, R. G.: Experimental ascites treated by hypophysectomy and substitution therapy, Quart. Bull. Northwestern Univ. M. School **32:** 201, 1959.
5. Davis, J. O., Goodkind, J. J., Pechet, M. M., and Ball, W. C.: Increased excretion of aldosterone in urine from dogs with right-sided congestive heart failure and from dogs with thoracic inferior vena cava constriction, Am. J. Physiol. **187:** 45, 1956.
6. Davis, J. O., Howell, D. S., and Southworth, J. L.: Mechanisms of fluid and electrolyte retention in experimental preparations in dogs. III. Effect of adrenalectomy and subsequent desoxycorticosterone acetate administration on ascites formation, Circulation Res. **1:** 260, 1953.
7. Davis, J. O., Yankopoulos, N. A., Liberman, F., Holman, J., and Bahn, R. C.: The role of the anterior pituitary in the control of aldosterone secretion in experimental secondary hyperaldosteronism, J. Clin. Invest. **39:** 765, 1960.
8. Hyatt, R. E., and Smith, J. R.: The mech-

anism of ascites: a physiological appraisal, Am. J. Med. **16:** 434, 1954.

9. Kreel, I., Canter, J. W., Segal, R. L., Frankel, A., and Baronofsky, I. D.: The influence of thyroidectomy on experimental ascites, S. Forum **10:** 125, 1960.
10. Leutscher, J. A., Jr., and Johnson, B. B.: Observations on sodium-retaining corticoid (aldosterone) in urine of children and adults in relation to sodium balance and edema, J. Clin. Invest. **33:** 1441, 1954.
11. McKee, F. W., Schilling, J. A., Tishkoff, G. H., and Hiatt, R. E.: Experimental ascites. Effects of sodium chloride and protein intake on protein metabolism of dogs with constricted inferior vena cava, Surg. Gynec. & Obst. **89:** 529, 1949.
12. Money, W. L.: The interrelation of the thyroid and the adrenals, Brookhaven Sympos. Biol. **7:** 137, 1955.
13. Peterson, R. E.: The influence of the thyroid on adrenal cortical function, J. Clin. Invest. **37:** 736, 1958.
14. Sprafka, J. L., Haddy, F. J., Alden, J. R., and Baronofsky, I. D.: The experimental production of tricuspid insufficiency and its relation to ascites, S. Forum **4:** 123, 1953.
15. Sprafka, J. L., Hannon, D. W., and Baronofsky, I. D.: The effect of subtotal adrenalectomy upon the development of ascites in chronic heart failure, S. Forum **5:** 143, 1954.
16. Veith, F. J., and Thrower, W. B.: A method of producing right-sided congestive heart failure in the dog, Surg. Gynec. & Obst. **109:** 687, 1959.
17. Yankopoulos, N. A., Davis, J. O., McFarland, J. A., and Holman, J.: Physiologic changes during chronic congestive heart failure in dogs with tricuspid insufficiency and pulmonic stenosis, Circulation Res. **7:** 950, 1959.

Effects of glycerol and freezing on skin homografts

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The purpose of this report is to demonstrate the effects of glycerol and of freezing on the "survival time" of mouse-skin homografts. The work of Brown[2] and Fryer[3] suggests that the viability of skin grafts is not significantly altered by freezing after the grafts have been treated with glycerol.

MATERIALS AND METHODS

White male mice* of an impure CF No. 1 strain were used so that all grafts would react antigenetically as homografts. The animals, 3 to 4 weeks of age, were divided into 3 groups. In Group I, 12 mice were grafted immediately with untreated skin, in Group II, 12 were grafted with skin treated with glycerol but not frozen, and in Group III, 12 were grafted with glycerol-treated frozen homografts.

Full-thickness sections of skin approximately 4 cm.^{2} in size were cut with scissors from the clipped ventral surfaces of donor animals. One portion of these sections was immediately secured to the control animals of Group I. The remaining grafts were placed on strips of sterile Reynolds aluminum foil coated with sterile mineral oil, with the raw dermal surface of the skin up, and immersed in a 15 per cent solution of glycerol (Harleco, 98 per cent) in lactated Ringer's solution (Abbott Laboratories). After remaining in this solution for 35 minutes the skin sections were removed and one section from each donor mouse was rinsed in plain lactated Ringer's solution and secured to a control mouse in Group II. The other sections of skin flaps were left on their aluminum-foil carriers, inserted into 18 by 160 mm. Pyrex test tubes, and immersed in a bath of solid carbon dioxide and ethanol at –72° C. for 3.5 hours. When removed from the bath, the test tube–graft assemblies were placed in a freezer at –28° C. One week later the test tubes were warmed to 37° C., and the foil carriers with skin were rinsed in 50 ml. of lactated Ringer's solution for 30 minutes. The skin was then secured to the dorsal surface of Group III mice.

All recipient animals were anesthetized with Nembutal for the operations. An area of skin approximately 3 mm.^{2} in size was cut from the dorsal surface of each recipient mouse. An incision was extended anteriorly and posteriorly to allow placement of a "Breedis chamber" frame.[1] After the insertion of this frame under the skin of the host animal, tab holes were cut for the frame and the incision was partially closed with No. 5-0 black silk. The skin graft was then placed on the defect to extend under the overlapping skin margins and the window of the Breedis chamber was wired on through the tabs (Fig. 1).

All mice were observed for indications of sloughing of the skin grafts. Biopsy specimens for microscopic study were taken from each group during various phases of the homograft rejection. Animals were given

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*Mice were donated by Baxter Laboratories, Inc., Cleveland, Miss.

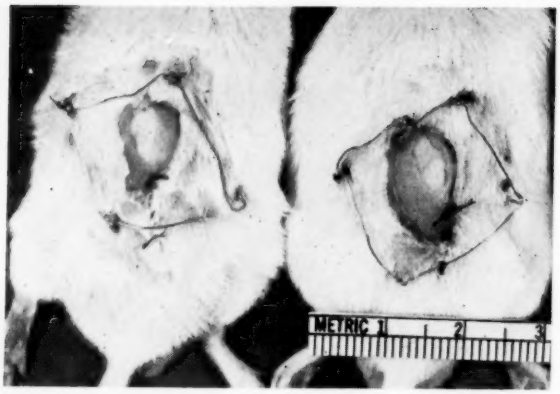

Fig. 1. Mice immediately after placement of homografts. Skin grafts are in place beneath the Breedis chamber frames.

Purina laboratory chow and tap water ad libitum.

RESULTS

In this work the survival period of a skin homograft is defined as the time between grafting and beginning of recession of the graft from its bed. Grafts on the control mice in Group I survived for an average of 15 days, plus or minus 1 day. Glycerol-treated grafts on mice in Group II survived 8 days, plus or minus 0.5 day, while the frozen glycerolized grafts on mice in Group III survived 18 days, plus or minus 1 day. Examination of microscopic slides made of the biopsy specimens showed the anticipated phases[5] of normal viability, acute leukocytic infiltration, edema, loss of vascular structure, and degeneration with necrosis of the epithelium. It is significant that these phases progressed rapidly in the glycerol-treated unfrozen grafts and corresponded with gross survival observations.

DISCUSSION

The survival periods of untreated grafts and of frozen grafts correspond to those reported by Brown.[2] However, neither Brown nor Fryer[3] reported employing a control graft treated with glycerol without freezing. The very short survival period of this type of graft seems to indicate that although glycerol is at present a sine qua non in the slow freezing of tissues, it is at the same time toxic in the concentrations used in this experiment in the temperature range of 20° to 37° C.

This hypothesis is strengthened by additional work in this laboratory and by the work of others. Meryman[6] has pointed out that "in many cases it is the glycerine itself which becomes toxic in concentrations sufficient to protect against freezing." Lovelock[4] has observed that red blood cells become hemolyzed in 1.5 M (13.8 per cent) glycerol at room temperature. In attempting to freeze rat hearts in glycerol solutions, we have found that if glycerol is perfused into the organs at room temperature in concentrations of 15 per cent the hearts cannot be resuscitated.[10] The concentration may be increased to this level only as the temperature drops. Possibly the "latent period" before the reappearance of the vaginal cycle in mice which received frozen ovarian grafts, as observed by Parks,[7] is due to the adverse effects of glycerol. Pomerat[8] has shown definitively with cinematic records the deleterious effect of glycerol on tissue cultures at 37° C. Concentrations as low as 10 per cent lead "to the separation of individual [cellular] elements at the periphery of the culture, but with the retention of narrow connecting strands which were optically dense. The cells . . . [showed] marked 'boiling' at the surface." At 20 per cent, "granules in the cytoplasm continued to show movements and boiling persisted." It must be noted, however, that even if glycerol does have a toxic effect on cells at body temperatures, this toxic effect may be tissue specific. Sherman[9] found that in human semen mixed 9 parts to 1 part absolute glycerol, glycerol pretreatment times could be as long as 30 minutes prior to freezing without significant effect on the survival of viable spermatozoa. Longer pretreatment periods were not studied.

SUMMARY

Full-thickness skin homografts were cut from the clipped ventral surface of donor mice and one portion immediately transplanted to a control group. Another portion

was temporarily placed in a 15 per cent solution of glycerol in lactated Ringer's solution, and then transplanted. A third portion of skin in the glycerol solution was frozen at –70° C., maintained at –28° C. for 1 week, thawed, and transplanted.

The immediately transplanted grafts survived 15 days before sloughing, grafts treated with glycerol, 8 days, and frozen grafts, 18 days.

CONCLUSION

Frozen glycerol-treated homografts in mice survive as long as immediately transplanted homografts. Grafts treated with 15 per cent glycerol at room temperature are adversely affected, presumably by the glycerol, with a resultant decrease in survival time.

REFERENCES

1. Breedis, C.: Regeneration of hair follicles and sebaceous glands from the epithelium of scars in the rabbit, Cancer Res. **14:** 575, 1954.
2. Brown, J. B., Fryer, M. P., Zaydon, T. J., and King, L.: Laboratory investigations of skin viability following preservation by various methods, S. Forum **6:** 577, 1956.
3. Fryer, M. P., and Brown, J. B.: The effect of various storage methods on the viability of skin grafts, Transplantation Bull. **3:** 96, 1956.
4. Lovelock, J. E.: Mechanism of the protective action of glycerol against haemolysis by freezing and thawing, Biochim. et biophys. acta **11:** 28, 1953.
5. Medawar, P. B.: The behaviour and fate of skin autografts and skin homografts in rabbits, J. Anat. **78:** 176, 1944.
6. Meryman, H. T.: Mechanics of freezing in living cells and tissues, Science **124:** 515, 1956.
7. Parks, A. S.: Viability of ovarian tissue after freezing, Proc. Roy. Soc., London, ser.B. **147:** 520, 1957.
8. Pomerat, C. M., and Moorhead, P. S.: Thermal stress and tissue culture. VII. The effect of glycerol on cells with special reference to protection against freezing, Texas Rep. Biol. & Med. **14:** 237, 1956.
9. Sherman, J. K.: Freezing and freeze-drying of human spermatozoa, Fertil. & Steril. **5:** 357, 1954.
10. Webb, W. R., and Karow, A. M.: Unpublished observations.

Isotope localization with tagged microspheres

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The introduction of chemotherapeutic agents into isolated areas of the body for treatment of malignant neoplasms has been partially successful and tumor growth has been controlled in some instances.[1] However, these techniques of perfusion generally have not been curative. The present investigations were undertaken to determine whether or not radioactive isotopes could be introduced into organs or regions of the body in a somewhat similar manner by the arterial route for the purpose of irradiation treatment of malignant tumors. In order to localize the isotope to the area involved and prevent its widespread diffusion throughout the body, it was considered necessary to introduce it in some physical form which would allow it to be trapped in the organ.

METHODS

To introduce the isotope into an organ in such manner that it was not distributed to the body in general, ceramic microspheres of uniform size (60 ± 5 microns) tagged with the isotope were utilized. These microspheres are small enough to embolize in the smaller arterioles and yet large enough that they do not pass through arteriovenous shunts to the general circulation in significant amounts. We believed that if such microspheres could be introduced into an organ in relatively small quantities, the number of vessels occluded would not be significant in terms of the circulation and nutrition of the organ, and infarction of the organ would not occur. The microspheres can be tagged with cationic isotopes and specific activities as great as 10^4 curies per gram of microspheres are obtainable. With high activity levels, only a few milligrams of the microspheres should be necessary in an organ to produce significant levels of internal irradiation.

The spheres have been introduced into organs of animals and several approaches have been made to study the effects.

Organ damage from embolization of the microspheres. Ceramic microspheres of the size described (diameter 60 ± 5 microns; density, 4; approximately 4,000 spheres per milligram), but isotope-free, were injected in varying amounts into the arteries which supply the kidney, liver, or lung in anesthetized adult mongrel dogs under aseptic conditions. The spheres were placed in a medium of 2 per cent gelatin, 50 per cent glucose, or 0.9 per cent saline for injection. A glass syringe and small-bore needle were used for injection. Ten days after the injection of the microspheres, animals were sacrificed and the organ removed and fixed with 10 per cent formalin. Sections were stained with hematoxylin and eosin.

Localization of microspheres in the organ and percentage loss into the general circulation. For these investigations the venous

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Table I. *Interarterial introduction of Y^{90}-laden microspheres*

Dog No.	Organ	Amount of spheres (mg.)	Total radio-activity (mc.)	Calculated radiation dosage* (rads)
1	Lung	15	3.9	10,800
2	Lung	20	8.0	14,200
3	Lung	15	3.9	8,300
4	Lung	36.1	14.4	22,000
5	Liver	40.1	16.0	6,800
6	Liver	22.3	8.9	4,000

*Assuming that 100 per cent of the spheres reached the organ.

outflow of liver, kidney, lung, or hind limb of dogs was collected by an inlying venous cannula during the period of intra-arterial injection of varying amounts of Cr^{51}-tagged microspheres and for a period of 5 minutes afterward. Animals were transfused with whole blood given into a peripheral vein during the period of venous bleeding to compensate for blood lost. Collected venous blood was digested with 10 per cent sodium hydroxide, filtered, and both the filter paper and an aliquot of the filtrate were counted in a well-type scintillation counter. Percentage of spheres lost into the general circulation could be calculated from these data.

Distribution of spheres in an organ as shown by autoradiography. A few hours after the introduction of microspheres tagged with tracer amounts of Y^{90} or Sm^{153} into the arteries supplying liver, lung, or kidney of mongrel dogs, the animals were sacrificed and the organ fixed with 10 per cent formalin. It was then sliced into one-fourth inch slices and autoradiographs of the organs made with ordinary x-ray film.

Effects of Y^{90}-laden spheres introduced into the liver and lung. For this experiment the isotope Y^{90} was attached to carrier microspheres of the same type and size utilized in previous experiments. Quantities of spheres varying in amount from 15 to 40.1 mg. with specific activities from 3.9 to 16 mc. were injected into the hepatic arteries or pulmonary arteries of dogs. Table I shows the amount of spheres and activity levels of Y^{90} administered to the various animals and the calculated radiation dosage to the organs involved. Animals were allowed to recover from the operative procedure. Peripheral blood studies, including determination of hemoglobin concentration, white cell count,

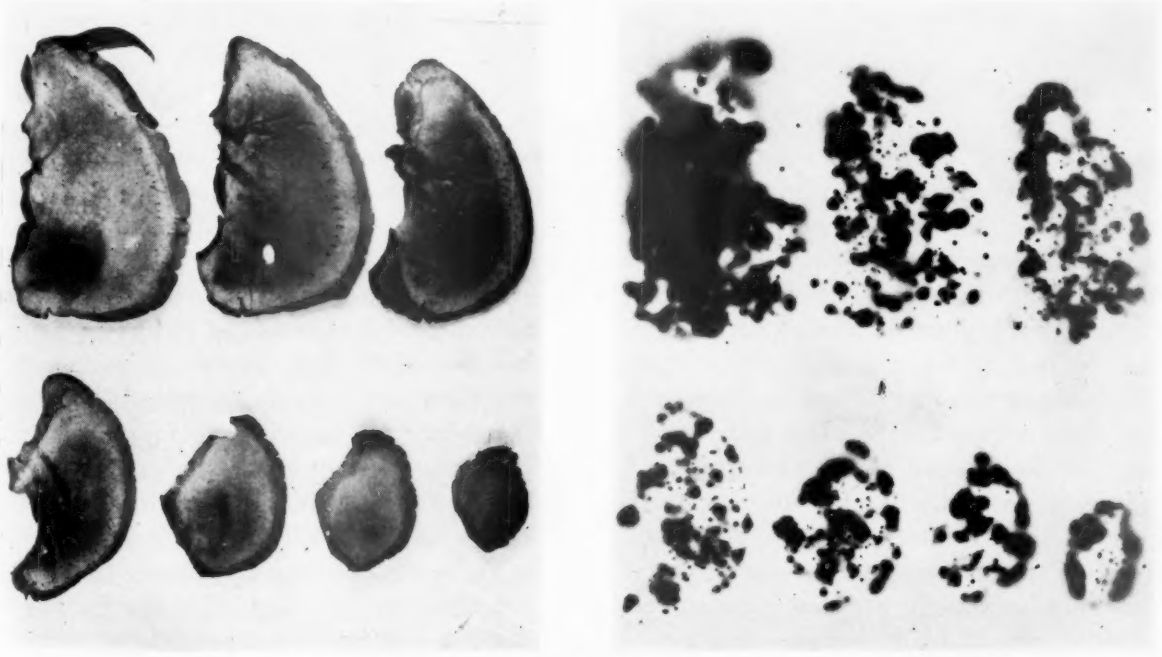

Fig. 1. Slices of formalin-fixed canine liver and corresponding autoradiograph.

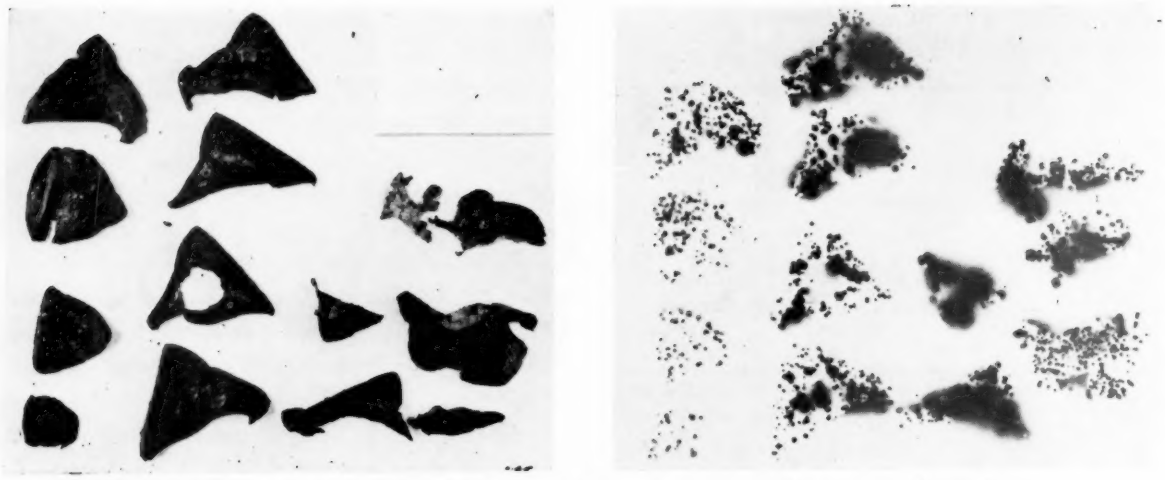

Fig. 2. Slices of formalin-fixed canine lung and corresponding autoradiograph.

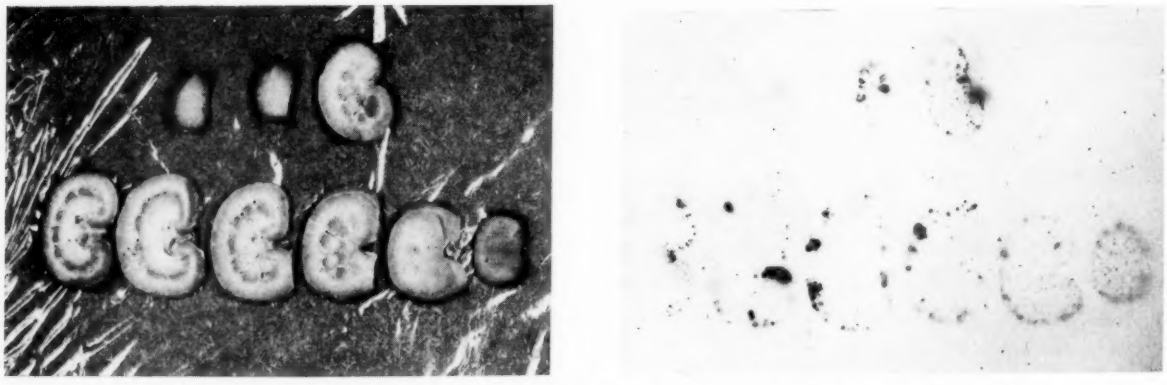

Fig. 3. Slices of formalin-fixed canine kidney and corresponding autoradiograph.

and differential were continued in animals for at least 48 days postoperatively. In animals in which the isotope had been injected into the hepatic artery, liver function was measured in the postinjection period by determination of serum bilirubin, alkaline phosphatase, and Bromsulphalein retention at intervals during the first 48 postoperative days. Preoperative liver function tests were carried out prior to injection of the spheres. Two of the dogs were sacrificed after the forty-eighth and sixty-ninth days in the postoperative period and the lungs removed. In the remaining animals liver or lung biopsies were taken at intervals in the postoperative period and the animals preserved for long-term observation. Histologic studies of the lungs and livers of these 6 animals will be reported.

RESULTS

Organ damage from embolization of the microspheres. Nine lobes of 3 canine lungs were studied 10 days after the intravascular introduction of 20 to 100 mg. of microspheres per lobe. All appeared grossly normal. Seven of the lobes showed no microscopic changes except minimal interstitial infiltration of round cells. The other 2 lobes from the same animal showed scattered areas of bronchopneumonia. Nine of 11 livers studied were grossly and microscopically normal after the injection of 20 to 150 mg. of

spheres into the hepatic arteries. Small areas of lobular necrosis in one liver and scattered areas of degeneration surrounding the central veins in another followed the introduction of 100 and 200 mg. of spheres, respectively, into the hepatic arteries of 2 dogs. Six of 11 kidneys showed severe cortical necrosis and the remaining 5 demonstrated lesser degrees of the same lesion after injection into the renal arteries of 20 to 100 mg. of microspheres.

Localization of microspheres in the organ and percentage loss into the general circulation. After injection of 9.6 to 13 mg. of Cr^{51}-tagged microspheres into the pulmonary arteries of 3 lungs, no more than 0.09 per cent passed through the lungs into pulmonary veins. The maximal recovery from femoral venous blood was 1 per cent of the total of 8.2 to 26.5 mg. of spheres injected into femoral arteries of dogs. After injection of 27 to 74.1 mg. of tagged spheres into the hepatic arteries of 3 animals, not more than 0.16 per cent were recovered from the suprahepatic inferior vena caval blood. Of 9.1 to 13.5 mg. of microspheres injected into the renal arteries of 3 animals, maximal recovery was 0.02 per cent of the total injected. Data on the 13 animals studied are summarized in Table II.

Distribution of spheres in an organ as shown by autoradiography. Distribution of microspheres in fixed tissue slices of liver and lung are shown in Figs. 1 and 2, respectively. Fairly uniform distribution throughout the small vessels of the organ occurred in all 5 livers and all 5 lungs studied. However, as can be seen in the autoradiographs, there is some tendency for more spheres to lodge centrally in the organ than in its periphery. This may be due to the relatively great density of the spheres. Autoradiographs of 3 kidneys showed nonuniform distribution with most of the spheres lodged in the cortex. Presumably they are blocked in transit at arterioles proximal to glomerular tufts and distribution throughout the medulla is prevented (Fig. 3).

Effects of Y^{90}-laden spheres introduced into the liver and lung. The 4 animals which

Table II. *Recovery of CR^{51}-tagged microspheres in venous blood after intra-arterial injection*

Animal No.	*Organ or region*	*Spheres injected (mg.)*	*% recovered in venous blood*
1	Lung	9.6	0.04
2	Lung	8.5	0.09
3	Lung	13.0	0.03
4	Kidney	13.4	0.01
5	Kidney	9.7	0.02
6	Kidney	9.1	0.00
7	Hind limb	19.4	0.20
8	Hind limb	10.5	1.00
9	Hind limb	26.5	0.15
10	Hind limb	8.2	0.84
11	Liver	45.9	0.16
12	Liver	74.1	0.14
13	Liver	27.0	0.04

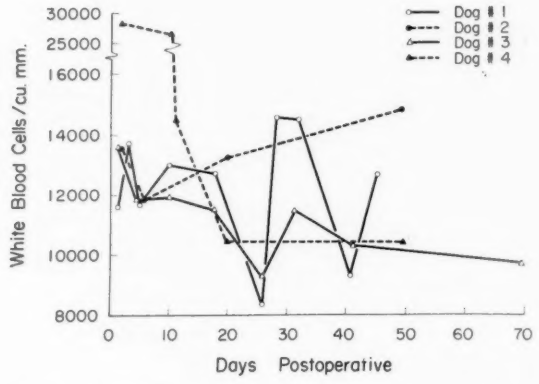

Fig. 4. Peripheral white blood cell counts after introduction of Y^{90}-laden microspheres into the canine pulmonary artery.

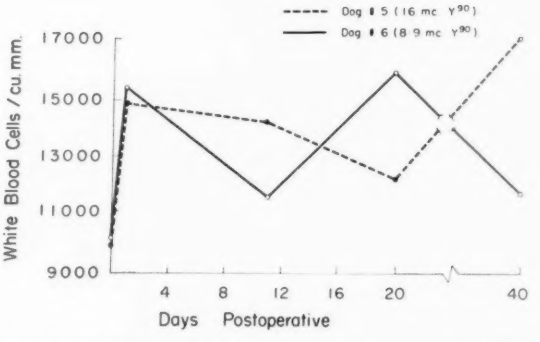

Fig. 5. Peripheral white blood cell counts after introduction of Y^{90}-laden microspheres into the canine hepatic artery.

received Y^{90}-laden spheres into the pulmonary artery and the 2 animals which received the isotope into the hepatic artery tolerated the procedure well and all survived for observation. Animals appeared healthy in the postoperative period, ate fairly well, and maintained their weight after an immediate postoperative weight drop. The leukocyte counts of animals studied can be seen in Figs. 4 and 5. It is apparent that no significant depression of white blood cell count occurred as a result of introduction of the spheres into the hepatic or pulmonary arteries. Similarly, hemoglobin values shown in Figs. 6 and 7 showed no great changes after introduction of the isotope into the pulmonary or hepatic arteries. The results of differential counts in all animals remained normal during the period of postoperative observation which extended for a period of at least 48 days in all animals studied. Serum bilirubin, alkaline phosphatase, and Bromsulphalein values are shown in Figs. 8, 9, and 10. There was a transient rise in the Bromsulphalein retention and serum alkaline phosphatase values after operation and introduction of the microspheres in each animal. These values returned to normal shortly thereafter and remained within normal limits for the 48-day observation period.

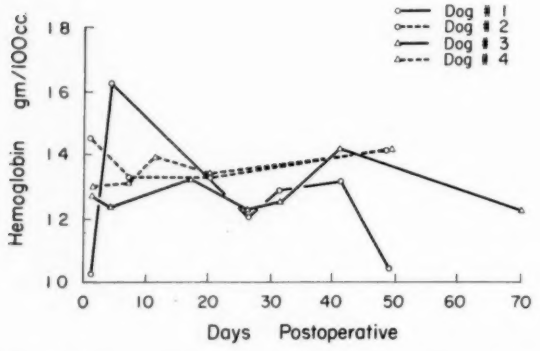

Fig. 6. Hemoglobin values after introduction of Y^{90}-laden microspheres into the canine pulmonary artery.

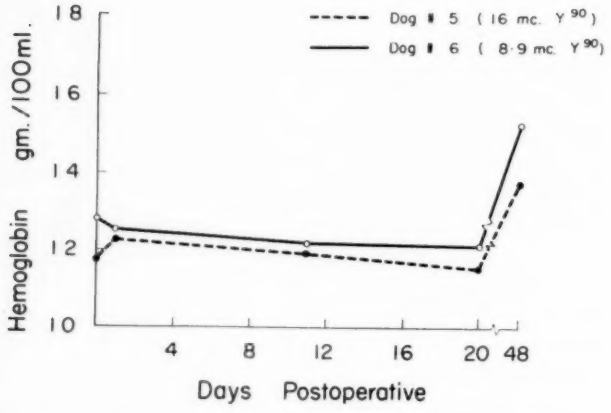

Fig. 7. Hemoglobin values after introduction of Y^{90}-laden microspheres into the canine hepatic artery.

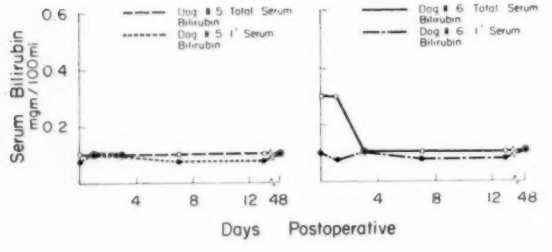

Fig. 8. Serum bilirubin values after introduction of Y^{90}-laden microspheres into the canine hepatic artery.

DISCUSSION

Experiments of Grim and Lindseth[2] demonstrated only 1 per cent of injected radioactive material to be found in the venous blood when spheres 43 microns in diameter were injected into the circulation of the intestine. Since a part, if not all, of the material in the venous blood was radiosodium washed out of the spheres, the authors concluded that all arteriovenous channels in this organ were less than 43 microns in diameter. They also found that only 4 per cent of spheres of 20 micron size passed into the venous outflow in the jejunum. The spheres with diameters in the 60 micron range used in our experiments were considered to be probably large enough to preclude their passage through most arteriovenous communications and yet sufficiently small to pass to the smaller arterioles. The results seem to support the conclusion that spheres of such sizes do behave in the manner predicted. Because the spheres are carried by arterial blood flow to all parts of an organ such as liver or lung, and because

they can be loaded with specific activities of radioisotopes sufficient to deliver therapeutic doses of irradiation to an organ, it should be possible to deliver intensive irradiation with only a few milligrams of spheres introduced by the arterial route. Microembolization of limited numbers of the spheres does not appear in itself to produce serious lesions in the organs studied, with the exception of the kidney. Additional animal experiments are in progress to investigate the effects of isotope-laden spheres with activities sufficient to deliver therapeutic dosages of irradiation. The results to date with Y^{90}-laden spheres for irradiation of liver and lung in dogs have been encouraging in that there has been no depression of the bone marrow and no significant alteration of liver function in animals studied. Measurement has shown that approximately 90 per cent of a given dose of microspheres can be delivered into the artery supplying an organ by the technique employed in these experiments. Therefore the calculated irradiation dosage to the organs of animals (Table I) is probably only slightly greater than that actually delivered.

To date 2 patients with metastatic deposits in the liver from colic cancer have been treated by this method. At operation approximately 3 and 4 mc. doses of Ba^{140} contained in 65 and 74 mg. of microspheres, respectively, were injected into the hepatic arteries of the 2 patients. Although the period of observation has been too short to determine the ultimate effect on the tumors, both patients have had uncomplicated postoperative courses and neither has demonstrated any significant change in liver function or peripheral blood count as demonstrated by conventional tests. Whether or not the technique has merit and what isotopes and irradiation dosages are optimal for use in patients can only be determined after more experience has been gained.

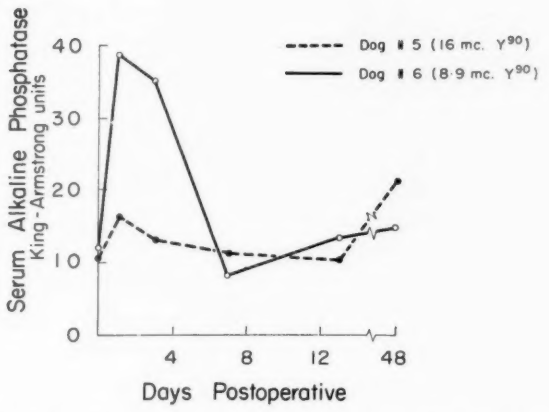

Fig. 9. Serum alkaline phosphatase levels after introduction of Y^{90}-laden microspheres into the canine hepatic artery.

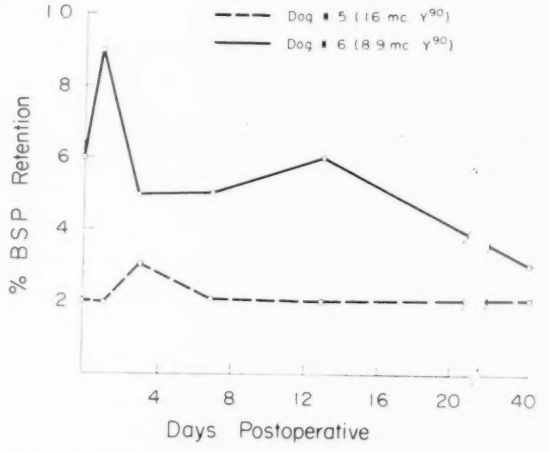

Fig. 10. Bromsulphalein retention after introduction of Y^{90}-laden microspheres into canine hepatic artery.

SUMMARY

1. Ceramic microspheres have been injected into the arteries of liver, kidney, lung, and hind limb of mongrel dogs and their localization and distribution studied.

2. The spheres themselves appear to cause very little damage by embolization in the organ studied when injected in small amounts, except in the kidney where they produce severe cortical necrosis. Autoradiographs made after introduction of isotope-tagged spheres showed fairly uniform random distribution throughout the organ.

3. Study of venous blood recovered during and after the injection of the spheres demonstrated no more than 1 per cent of such spheres to pass through arteriovenous communications into the general circulation in any experiment.

4. In 6 animals experiments with micro-

spheres with activity levels of Y^{90} varying from 3.9 to 16 mc. demonstrated no depression of the blood elements and no significant interference with liver function.

5. Injection of isotope-laden microspheres into the hepatic arteries of 2 patients with metastatic liver deposits from colic cancer has resulted in no demonstrable unfavorable effects. The patients are currently being studied to determine ultimate effects on their tumors.

The authors wish to express their appreciation to Dr. James R. Johnson, Dr. John P. Ryan, Mr. T. N. Lahr, and Mr. L. E. LaMar of the Nuclear Products Research Section of Minnesota Mining and Manufacturing Company, who prepared the microspheres used in these experiments (3m brand radiating microspheres).

REFERENCES

1. Creech, O., Krementz, E. T., Ryan, R. F., Reemtsma, K., and Winblad, J. N.: Experiences with isolation-perfusion technics in the treatment of cancer, Ann. Surg. **149:** 627, 1959.
2. Grim, E., and Lindseth, E. O.: Distribution of blood flow to the tissues of the small intestine of the dog, Univ. Minnesota M. Bull. **30:** 138, 1958.

Evaluation of Warexin* as a potential cold sterilizing agent

I. BACTERIOLOGIC STUDIES

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The development of epidemiologic problems pertaining to resistant strains of Staphylococcus have reawakened our concern for safe cold sterilization methods. Many different chemicals have been recommended and are effective. However, certain chemicals, because of their strong oxidizing, corrosive, or toxic action, cannot be used. Others require long time periods of contact with the bacteria, and for some, special, complex, bulky equipment must be available.

The purpose of this study is twofold, first, to describe a new protocol for the evaluation of cold sterilizing agents and, second, to report the results with a chemical agent, Warexin, tested by this procedure. This monoxychlorosene derivative[1, 2, 4-8] contains an organic hypochlorous acid in a phosphate buffer. It is a white powder with a characteristic chlorine odor and dissolves readily in distilled water or in soft tap water (100 ppm hardness or less). There is a period of induction in its solubility curve, that is, the product rehydrates prior to dissolving. A 1.25 per cent solution of Warexin in distilled water has a pH range of 7.4 to 7.8. It is noncorrosive, destroys microorganisms within a reasonable time limit, and requires very simple laboratory materials to provide safe cold sterilization.

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*"Warexin" is the registered trade mark of the Guardian Chemical Corporation, Long Island City, N. Y., for buffered monoxychlorosene. It is distributed by Davol Rubber Company, Providence, R. I.

MATERIALS AND METHODS

Antimicrobial action of Warexin at varied concentrations and different time intervals. Warexin concentrations of 2, 1.5, 1.25, 1.00, and 0.5 per cent were prepared in 500 ml. of tap water in clean, nonsterile gallon Duraglass jars or 2 L. beakers. The mixtures were stirred or shaken repeatedly at 10-minute intervals until complete dissolution occurred. The control consisted of a sixth 500 ml. jar containing only tap water. Five milliliter amounts of a bacterial suspension were added to each jar and at different time intervals 5 ml. aliquots were removed, transferred to sterile test tubes, and then streaked with cotton swabs onto plated, solid-surface media.

The bacteria tested included aerobic and anaerobic spore formers, a phage-sensitive (42B/52/47C/80/81) Staphylococcus which was resistant to penicillin, streptomycin, and the tetracyclines (a hospital epidemic strain), an encapsulated bacterium, a gram-negative rod, and an acid-fast organism.

Bacillus stearothermophilus was grown on 3 trypticase agar flats (growth media). The flats were prepared with 250 ml. prescription bottles, heavily inoculated, and incubated at 65° C. for 24 to 48 hours. Fifteen milliliters of sterile saline was then added to each flat. Growth was removed from the solid surface by simple rotation and/or scraping with an inoculating loop. The contents of the 3 flats were pooled. A Gram stain was made to demonstrate purity of culture and 10 ml. aliquots

were removed for turbidimetric reading on the Coleman Junior 6A spectrophotometer with the 650μ wave length. The percentage readings were converted to organisms per milliliter on a standard conversion table.[3] The range for all bacteria tested numbered between 2,100 million and 3,900 million per milliliter. One milliliter of the bacterial suspension was then added to each of 3 sterile empty Petri dishes and incubated at 65° C. At weekly intervals, for a total of 3 weeks, trypticase agar was added to determine spore survival. Five milliliter aliquots of the pooled bacterial suspension were then added to each of the 6 jars containing the different concentrations of Warexin and the water control. After time intervals of 1, 2.5, 5, 15, 30, and 60 minutes, 5 ml. aliquots were removed and added to sterile test tubes immersed in an ice bath. Sterile cotton swabs were then introduced into each 5 ml. aliquot and streaked onto a divided trypticase agar plate (plating media). When all segments were streaked, the plates were incubated, in this case at 65° C. for 48 hours. Growth or absence of growth was recorded. With this method a total of 6 plates is used (2, 1.5, 1.25, 1, and 0.5 per cent and water control) for each organism tested. Each plate was divided into 8 segments, 6 to use for the different time intervals, 1 for the cotton swab streak of organisms not exposed to Warexin or tap water, and, finally, 1 segment to serve as the uninoculated control to insure sterility of the trypticase agar plate. *Bacillus subtilis, Staphylococcus pyogenes* var. *aureus, Klebsiella pneumoniae,* and *Pseudomonas aeruginosa* were grown, prepared, and tested in the same manner, with the exception that nutrient agar flats were used for growth and plating media, and an incubation temperature of 37° C. for 24 to 48 hours was used.

Dubos Tween 80 agar for plating media and Dubos Tween 80 liquid for growth media were used for *Mycobacterium tuberculosis* var. *hominis.* An incubation temperature of 37° C. for 4 to 6 weeks was used. To prevent drying, the Petri dishes were sealed with wide rubber bands.

The procedure described was modified for the Clostridia. Thioglycollate broth was used for the growth media and pooled to obtain the desired turbidity. The "plating media" also consisted of thioglycollate broth. In the latter instance the entire swab was introduced and remained in the tube during the 48-hour incubation period.

Results. Exposure of 6 of the 8 bacteria (*Bacillus subtilis, Clostridium perfringens, Staphylococcus pyogenes* var. *aureus, Klebsiella pneumoniae, Pseudomonas aeruginosa,* and *Mycobacterium tuberculosis* var. *hominis*) for 1 minute in the 5 different concentrations of Warexin caused complete killing. One bacterium, *Bacillus stearothermophilus,* survived for 2.5 minutes. However, this occurred at the lower concentrations of Warexin (1 and 0.5 per cent). The 1-minute exposure failed to destroy *Clostridium sporogenes* at any of the concentrations used.The longer time intervals resulted in effective cold sterilization. The water control showed growth on all segments except, of course, the uninoculated. The spore formers, the encapsulated bacterium, and the acid-fast organism survived drying for 3 weeks. Staphylococcus and Pseudomonas resisted desiccation for 2 weeks. Our over-all results indicate that when these organisms, ranging in number of 2,100 million to 3,900 million per milliliter, remain in contact with Warexin for at least 5 minutes, any range between 0.5 to 2.0 per cent concentration is effective for cold sterilization.

Antimicrobial action of Warexin in the presence of organic material. This experiment, with one exception, was identical to the first. In this study each jar of Warexin and control contained 5 per cent human serum.

There appeared to be a definite optimal destructive range for each bacterium tested, that is, a concentration of Warexin plus serum that killed the organism within 30 seconds or 1 minute. The aerobic spore former, *Bacillus subtilis,* failed to survive the 1 per cent Warexin preparation containing 5 per cent serum, while 60 minutes was necessary to destroy it in the 2, 3, and 4 per cent concentrations. The 5 per cent

serum-Warexin concentration killed this organism within 15 minutes. The optimal killing range for *Bacillus stearothermophilus* was 2 or 3 per cent, while 30, 15, and 5 minutes were required for destruction of this organism at 1, 4, and 5 per cent concentrations, respectively. The anaerobic spore formers *Clostridium sporogenes* and *perfringens* showed an optimal range of 4 and 3 per cent, respectively. Furthermore, a 30-minute exposure was required to destroy *Clostridium sporogenes* at the 1, 2, and 3 per cent levels, while 15 minutes sterilized the preparation with 5 per cent Warexin. For *Clostridium perfringens*, 30 minutes was required at the 1 per cent level, and 15 minutes at the 4 and 5 per cent concentrations. *Klebsiella pneumoniae* had an optimum of 3 per cent, while a 15-minute exposure was sufficient at the 5, 4, and 2 per cent levels, and 30 minutes at the 1 per cent concentration. The Pseudomonas species showed 3 optimal killing ranges, 5, 4, and 2 per cent, the 3 and 1 per cent concentrations required 15 and 60 minutes, respectively, to destroy the organism. The Staphylococci were readily killed within 5 minutes in the 5, 4, 3, and 2 per cent mixtures, while 60 minutes was required with the 1 per cent. Higher levels of Warexin, 5 and 4 per cent, were required to destroy the Mycobacteria within 15 minutes. Thirty and 60 minutes were required for the lower Warexin concentrations. With the different sera containing Warexin concentrations it is evident that for bacterial concentrations ranging from 1,150 million to 3,600 million organisms per milliliter, 60 minutes' contact should be maintained to insure effective cold sterilization. Whether these optima change or persist with larger concentrations of sera remains to be elucidated. All 8 bacteria soaked in serum survived desiccation 3 weeks.

Antimicrobial action of Warexin with contaminated materials immersed in different concentrations and for varying intervals of time. A protocol was developed to simulate testing of contaminated instruments by the cold sterilization technique. For "test" substances we chose 2 inch stainless steel screws, dental burrs of all sizes, and surgical Marlex mesh cut into 1 inch squares. These materials provided adequate surface areas along with points of lodgement for the bacteria.

All Warexin concentrations (without serum) and bacterial suspensions were prepared as before. However, instead of adding 5 ml. of the pooled bacterial suspension in each jar, we added the entire pooled amount to a large sterile Petri dish or beaker containing 36 stainless steel screws, 36 dental burrs, and 36 pieces of mesh. These materials were soaked for 30 minutes, after which time 6 of each of the contaminated items were added to each Warexin concentration and control jar. At intervals of 30 seconds, 1.5, 5, 15, 30, and 60 minutes, one of each of the objects was removed from the jars and dropped separately into the growth media tubes, incubated at the optimum growth temperature, and observed after 48 hours.

Nutrient broth (with phenol red indicator) was used as the medium for the screws, burrs, and mesh soaked with Staphylococci, Klebsiella, Pseudomonas and the Subtilis species of bacillus. The incubation temperature was 37° C. Similar items soaked with the 2 species of Clostridium were added to thioglycollate broth and incubated at the same temperature. Materials soaked with *Bacillus stearothermophilus* and *Mycobacterium tuberculosis* var. *hominis* were introduced into trypticase broth and Dubos Tween 80 liquid media, respectively. *Bacillus stearothermophilus* was incubated at 65° C. and the Mycobacterium at 37° C. It should be noted that trace amounts of turbidity could be caused by other substances carried over with the object tested. Furthermore, at this stage we did not know whether lack of turbidity or merely a trace of cloudiness reflected a true killing effect (bacteriolysis) or merely an inhibitory effect (bacteriostasis). To answer this question a plate had to be streaked from the contents of each subculture tube containing the screw, burr, and mesh. These plates were divided into 6 segments, each segment representing a different time, and each plate a different

Table I. *Survival time of bacteria soaked onto dental burrs, Marlex mesh, and stainless steel screws at different concentrations of Warexin and varied time intervals*

Bacteria	Optical density at wave length 650 μ and No. of organisms/ml.	2.0% Warexin						1.5% Warexin						1.25% Warexin						1.0% Warexin						0.5% Warexin						Water control						Growth after desiccation, 37° C. (wk.)		
		1	2	3	4	5	6	1	2	3	4	5	6	1	2	3	4	5	6	1	2	3	4	5	6	1	2	3	4	5	6	1	2	3	4	5	6	1	2	3
Bacillus subtilis	72.5% 1,150 M.																																					+	+	+
Tube																																								
Mesh		+	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	+	−	−	−	−	−	+	+	+	−	−	−	+	+	+	+	+	+	+	+	+
Screws		−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	+	+	−	−	−	−	+	+	+	−	−	−	+	+	+	+	+	+	+	+	+
Burrs		+	+	−	−	−	−	+	+	+	−	−	−	+	+	+	−	−	−	+	+	+	+	−	−	+	+	+	+	−	−	+	+	+	+	+	+	+	+	+
Plate																																								
Mesh		−	−	−	−	−	−	+	−	−	−	−	−	+	−	−	−	−	−	+	+	+	−	−	−	+	+	+	−	−	−	+	+	+	+	+	+			
Screws		−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	+	+	+	+	+	+			
Burrs		+	+	+	−	−	−	+	+	+	−	−	−	+	+	+	+	−	−	+	+	+	+	−	−	+	+	+	+	+	−	+	+	+	+	+	+			
Klebsiella pneumoniae	51% 2,200 M.																																					+	+	+
Tube																																								
Mesh		−	−	−	−	−	−	−	−	−	−	−	−	+	−	−	−	−	−	+	+	−	−	−	−	+	+	−	−	−	−	+	+	+	+	+	+	+	+	+
Screws		+	+	−	−	−	−	+	+	+	−	−	−	+	+	+	−	−	−	+	+	+	−	−	−	+	+	+	+	−	−	+	+	+	+	+	+	+	+	+
Burrs		+	−	−	−	−	−	+	+	+	−	−	−	+	+	+	−	−	−	+	+	+	−	−	−	+	+	+	+	−	−	+	+	+	+	+	+	+	+	+
Plate																																								
Mesh		−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	+	−	−	−	−	−	+	+	−	−	−	−	+	+	+	+	+	+			
Screws		−	−	−	−	−	−	−	−	−	−	−	−	+	−	−	−	−	−	−	−	−	−	−	−	+	+	+	−	−	−	+	+	+	+	+	+			
Burrs		+	+	−	−	−	−	+	+	+	−	−	−	+	+	+	−	−	−	+	+	+	−	−	−	+	+	+	+	−	−	+	+	+	+	+	+			
Pseudomonas aeruginosa	18% 6,600 M.																																					+	+	−
Tube																																								
Mesh		−	−	−	−	−	−	−	−	−	−	−	−	+	−	−	−	−	−	+	+	−	−	−	−	+	+	−	−	−	−	+	+	+	+	+	+	+	+	+
Screws		+	−	−	−	−	−	+	−	−	−	−	−	−	−	−	−	−	−	+	−	−	−	−	−	+	+	+	+	−	−	+	+	+	+	+	+	+	+	+
Burrs		+	+	−	−	−	−	+	+	−	−	−	−	+	+	+	+	−	−	+	+	+	+	−	−	+	+	+	+	−	−	+	+	+	+	+	+	+	+	+
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Mesh		−	−	−	−	−	−	−	−	−	−	−	−	+	−	−	−	−	−	+	+	−	−	−	−	+	+	+	−	−	−	+	+	+	+	+	+			
Screws		+	−	−	−	−	−	+	−	−	−	−	−	+	−	−	−	−	−	+	+	+	−	−	−	+	+	+	−	−	−	+	+	+	+	+	+			
Burrs		+	+	−	−	−	−	+	+	−	−	−	−	+	+	+	+	−	−	+	+	+	+	−	−	+	+	+	+	−	−	+	+	+	+	+	+			
Staphylococcus pyogenes var. *aureus*	45% 2,550 M.																																					+	+	−
Tube																																								
Mesh		−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	+	−	−	−	−	−	+	−	−	−	−	−	+	+	+	+	+	+	+	+	+
Screws		+	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	+	−	−	−	−	−	+	+	−	−	−	−	+	+	+	+	+	+	+	+	−
Burrs		+	+	+	−	−	−	+	+	+	−	−	−	+	+	+	−	−	−	+	+	+	+	−	−	+	+	+	+	−	−	+	+	+	+	+	+	+	+	+
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Screws		−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	+	−	−	−	−	−	+	+	+	−	−	−	+	+	+	+	+			
Burrs		+	+	+	−	−	−	+	+	+	−	−	−	+	+	+	−	−	−	+	+	+	+	−	−	+	+	+	+	+	−	+	+	+	+	+			
Bacillus stearothermophilus	37% 3,250 M.																																				+	+	+
Tube																																							
Mesh		−	−	−	−	−	−	+	+	−	−	−	−	+	+	−	−	−	−	+	+	+	−	−	−	+	+	+	+	−	−	+	+	+	+	+	+	+	+
Screws		−	−	−	−	−	−	−	−	−	−	−	−	+	−	−	−	−	−	+	+	−	−	−	−	+	+	+	+	−	−	+	+	+	+	+	+	+	+
Burrs		+	+	−	−	−	−	+	+	−	−	−	−	+	+	+	+	−	−	+	+	+	+	−	−	+	+	+	+	+	−	+	+	+	+	+	+	+	+
Plate																																							
Mesh		+	−	−	−	−	−	+	+	−	−	−	−	+	+	−	+?	−	−	+	+	+	+	−	−	+	+	+	+	+	−	+	+	+	+	+			
Screws		+	−	−	−	−	−	+	−	−	−	−	−	+	−	−	−	−	−	+	−	+	−	−	−	+	+	+	+	+	−	+	+	+	+	+			
Burrs		+	+	−	−	−	−	+	+	−	−	−	−	+	+	+	−	−	−	+	+	+	+	−	−	+	+	+	+	+	−	+	+	+	+	+			
Clostridium perfringens	69% 1,275 M.																																				+	+	+
Tube																																							
Mesh		−	−	−	−	−	−	−	−	−	−	−	−	+	+	−	−	−	−	+	+	−	−	−	−	+	+	+	−	−	−	+	+	+	+	+	+	+	+
Screws		+	−	−	−	−	−	+	+	−	−	−	−	+	+	+	−	−	−	+	+	+	+	−	−	+	+	+	+	−	−	+	+	+	+	+	+	+	+
Burrs		+	+	+	−	−	−	+	+	+	−	−	−	+	+	+	+	−	−	+	+	+	+	−	−	+	+	+	+	−	−	+	+	+	+	+	+	+	+
Plate																																							
Mesh		−	−	−	−	−	−	+	−	−	−	−	−	+	+	−	−	−	−	+	+	−	−	−	−	+	+	+	+	−	−	+	+	+	+	+			
Screws		+	−	−	−	−	−	+	+	−	−	−	−	+	+	+	−	−	−	+	+	+	−	−	−	+	+	+	+	−	−	+	+	+	+	+			
Burrs		+	+	+	−	−	−	+	+	+	−	−	−	+	+	+	+	−	−	+	+	+	−	−	−	+	+	+	+	−	−	+	+	+	+	+			
Clostridium sporogenes	38% 3,000 M.																																				+	+	+
Tube																																							
Mesh		+	−	−	−	−	−	+	+	−	−	−	−	+	+	−	−	−	−	+	+	+	−	−	−	+	+	+	+	+	−	+	+	+	+	+	+	+	+
Screws		+	−	−	−	−	−	+	−	−	−	−	−	+	−	−	−	−	−	+	+	−	−	−	−	+	+	+	+	−	−	+	+	+	+	+	+	+	+
Burrs		+	+	−	−	−	−	+	+	−	−	−	−	+	+	+	−	−	−	+	+	+	−	−	−	+	+	+	+	−	−	+	+	+	+	+	+	+	+
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Mesh		+	−	−	−	−	−	+	−	−	−	−	−	+	+	−	−	−	−	+	+	−	−	−	−	+	+	+	−	+	−	+	+	+	+	+			
Screws		+	−	−	−	−	−	+	+	−	−	−	−	+	−	−	−	−	−	+	+	−	−	−	−	+	+	+	+	+	−	+	+	+	+	+			
Burrs		+	−	−	−	−	−	+	+	−	−	−	−	+	+	+	−	−	−	+	+	+	+	−	−	+	+	+	+	−	−	+	+	+	+	+			
Mycobacterium tuberculosis* var. *hominis	40% 2,900 M.																																				+	+	+
Tube																																							
Mesh		+	−	−	−	−	−	+	−	−	−	−	−	+	−	−	−	−	−	+	+	+	−	−	−	+	+	+	±	−	−	+	+	+	+	+	+	+	+
Screws		+	+	−	−	−	−	+	+	−	−	−	−	+	+	−	−	−	−	+	+	+	−	−	−	+	+	+	−	−	−	+	+	+	+	+	+	+	+
Burrs		+	+	−	−	−	−	+	+	−	−	−	−	+	+	+	−	−	−	+	+	+	−	−	−	+	+	+	+	−	−	+	+	+	+	+	+	+	+
Plate																																							
Mesh		+	−	−	−	−	−	+	−	−	−	−	−	+	−	−	−	−	−	+	+	+	−	−	−	+	+	+	−	−	−	+	+	+	+	+			
Screws		+	+	−	−	−	−	+	+	−	−	−	−	+	+	−	−	−	−	+	+	+	−	−	−	+	+	+	+	−	−	+	+	+	+	+			
Burrs		+	−	−	−	−	−	+	−	−	−	−	−	+	+	−	−	−	−	+	+	+	−	−	−	+	+	+	+	−	−	+	+	+	+	+			

+ = Growth.
− = No growth.
6 = Item removed after 60 minutes in Warexin.
5 = Item removed after 30 minutes in Warexin.
4 = Item removed after 15 minutes in Warexin.
3 = Item removed after 5 minutes in Warexin.
2 = Item removed after 2.5 minutes in Warexin.
1 = Item removed after 30 seconds in Warexin.

concentration of Warexin. Cotton swabs were introduced into each tube and streaked onto the appropriate segment of the plate. Temperatures of incubation and media were the same as those described for the broth cultures.

In Table I growth or absence of growth is recorded for the screws, dental burrs, and surgical mesh which were soaked in the 8 different bacteria, exposed for varied periods of time in the different concentrations of Warexin, and finally added to broth and then streaked onto plates. The broth observations are recorded as tube results, and the plates as plate results.

If we consider all concentrations of Warexin tested, it is evident that if the mesh, screws, and burrs contaminated with 1,150 million to 6,600 million organisms per milliliter maintained contact for 30 to 60 minutes, the organisms are completely destroyed. For the higher concentration of disinfectant, however, less time was required. The one exception was *Clostridium sporogenes,* which required the full 30 to 60 minutes to destroy. The 30-second time interval in most instances was not sufficient for any of the agents tested. In general, the bacteria-soaked dental burrs resisted destruction over a longer time period, generally 5 minutes. In this regard it should be kept in mind that the dental burrs were not brushed clean before they were added to the disinfectant. We merely collected old, used, discarded dental burrs from clinics and used them as provided. Moreover, the Warexin solution was not agitated after the addition of the screws, bolts, and mesh. The results indicate Warexin to be a satisfactory cold sterilizing agent. In general, exposure of 2.5 minutes, and definitely 5 minutes, killed all the organisms tested at one of the concentrations used.

The bacteria on the soaked objects resisted desiccation for the 3-week testing period with the exception of the Pseudomonas and the Staphylococci which lost viability after the second week.

SUMMARY

A new procedure was introduced for the evaluation of cold sterilizing agents. Eight different bacteria prepared separately either as pooled preparations, as pooled preparations soaked onto 2-inch stainless steel screws, dental burrs, and Marlex mesh, or as pooled preparations added to 5 per cent serum mixtures, were added to varied concentrations of Warexin. Aliquots were removed at different time intervals and tested for bacterial survival. Results indicated Warexin to possess, under all 3 experimental conditions, effective sterilizing action within the set time limits.

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REFERENCES

1. Allen, J. G.: Extracorporeal circulation, Springfield, Ill., 1958, Charles C Thomas, Publisher, p. 87.
2. Gliedman, M., Grant, R., Vestal, B., Rogers, C., and Kallson, K. E.: Clorpactin, a surgical adjunct: antimicrobial and tumorcidal properties, S. Forum, vol. 3, 1958.
3. Gradwohl, R. B. H.: Clinical laboratory methods and diagnosis, St. Louis, 1956, The C. V. Mosby Company, ed. 5, vol. 2, p. 1477.
4. Lattimer, G. K., and Spirito, A. L.: Clorpactin for tuberculous cystitis, J. Urol. **73:** 1015-1018, 1955.
5. Paulo de Almeida, F., et al.: Action of Clorpactin WCS-90 in microbiology, Rev. paulista med. **43:** 104-105, 1953.
6. Swanker, W. A.: Use of Clorpactin WCS-90 as an antiseptic in surgery, Am. J. Surg. **90:** 44-46, 1955.
7. Wolinsky, E., Smith, M. M., and Steenken, Wm., Jr.: Tuberculocidal activity of Clorpactin, a new chlorine compound, Antibiotic Medicine, **1:** 382-384, 1955.
8. Zwelling, M.: Clorpactin WCS-90: a new antiseptic, A. M. A. Arch. Otolaryng. **62:** 157-158, 1955.

Physiologic considerations in the management of patients with corticosteroid-induced adrenal suppression who undergo operation

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The importance of an intact adrenal cortex in an organism's ability to withstand an acute stress is well illustrated by the inability of the adrenalectomized animal to survive surgical trauma and by the occurrence of sudden vascular collapse and sometimes death after operation in patients with adrenocortical atrophy secondary to prolonged corticosteroid therapy.[5, 16, 17] Pharmaceutical firms have been successful in the past decade in developing chemical analogues of either cortisol or cortisone which have the therapeutic advantage of being powerful anti-inflammatory agents and yet do not cause the objectionable "side effects" of sodium retention and potassium loss.[7, 19] Unfortunately, as each new synthetic hormone became commercially available, the clinician was presented with a steroid that was more effective than its predecessor in its ability to produce adrenocortical atrophy.[3, 8] Dexamethasone, the most recently introduced cortisol analogue, has been shown to be 700 times as potent as cortisol in producing adrenal atrophy in the rat and possesses at least 75 times the potency of cortisol in suppressing corticotropin secretion in man.[1]

In view of the widespread use of the newer cortisol analogues, it is essential for the clinician to have an understanding of the physiologic principles involved in carrying a patient who has been on prolonged corticosteroid therapy prior to operation safely through a surgical procedure. It is the purpose of this paper to report our clinical and hormonal observations on a group of such patients who were subjected to varying degrees of surgical stress.

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METHODS AND MATERIALS

Thirteen patients who ranged in age from 27 to 68 years were selected so that the series would include patients who underwent major and minor elective surgical procedures. There was only 1 woman in the group. One subject had undergone a bilateral total adrenalectomy for metastatic carcinoma of the prostate 10 months before this study. The majority of operations were performed under ether or cyclopropane anesthesia. Spinal anesthesia was employed in 5 cases.

All patients were receiving corticosteroid therapy immediately prior to the day of operation except for 1 individual for whom prednisone therapy had been discontinued 1 week before operation (Table I). The adrenocortical response to a 6-hour saline infusion of 30 I.U. of ACTH was measured 1 to 7 days preoperatively in 12 patients. Hormone therapy was discontinued for 24 hours before the performance of the ACTH

Table I. *Maintenance hormone therapy for medical disease prior to operation*

Subject	*Age*	*Disease*	*Hormone*	*Range of daily dosage (mg.)*	*Duration*
J. K.	64	Bronchial asthma	Triamcinolone	2-24	7 mo.
J. G.	42	Bronchial asthma	Cortisone	100-200	4 yr.
			Prednisolone	20	12 mo.
R. L.	63	Bronchial asthma	Dexamethasone	3-5	5 mo.
			Prednisone	20	5 wk.
H. D.	27	Ulcerative colitis	Prednisone	10-20	5 wk.
P. D.	62	Rheumatoid arthritis	Prednisolone	15-20	3 yr.
B. O.	29	Ulcerative colitis	Prednisone	5-60	6 mo.
				0	1 wk.
L. F.	68	Neurodermatitis	Cortisone	75-150	6 yr.
			6-Methylprednisolone	6-8	4 mo.
J. F.	64	Rheumatoid arthritis	Cortisone	50-150	5 yr.
			Prednisone	10-15	4 mo.
H. C.	58	Rheumatoid arthritis	Prednisone	10-30	3 yr.
			Dexamethasone	0.75-3	6 mo.
F. W.	48	Neurodermatitis	Cortisone	100-300	7 yr.
			6-Methylprednisolone	24	3 mo.
E. V.	33	Rheumatoid arthritis	Triamcinolone	6-8	6 mo.
E. D.	63	Rheumatoid arthritis	Cortisone	50-100	4 yr.
			Prednisone	10-20	1 yr.
E. B.	65	Addison's disease	Cortisone	50	10 mo.
			Fluorocortisol	2	1 wk.

Table II. *Supplementary hormone therapy for surgical operation*

	Evening before operation (8 to 10 P.M.)		*Morning before operation (6 to 7 A.M.)*		*Immediate postoperative period (2 to 8 P.M.)*	
Subject	*Hormone*	*Dose (mg.)*	*Hormone*	*Dose (mg.)*	*Hormone**	*Dose (mg.)*
J. K.	Fluorocortisol	4	Fluorocortisol	2	Cortisone	100
	Fluorocortisol	4	Fluorocortisol	2	Cortisone	100
J. G.	Fluorocortisol	4	Fluorocortisol	2	Cortisone	100
R. L.	Fluorocortisol	4	Fluorocortisol	2	Cortisone	100
H. D.	Fluorocortisol	4	Fluorocortisol	2	Cortisone	100
P. D.	Dexamethasone	1.5	Dexamethasone	3	Dexamethasone	3
B. O.	Dexamethasone	3	Dexamethasone	3	Cortisone	100
L. F.	Triamcinolone	4	Triamcinolone	8	Triamcinolone	8
J. F.	Triamcinolone	8	Triamcinolone	12	Triamcinolone	12
H. C.	Triamcinolone	8	Triamcinolone	16	Triamcinolone	24
F. W.	—	0	—	0	Prednisolone	50
E. V.	—	0	—	0	Prednisolone	50
E. D.	Prednisolone	50	—	0	Cortisone	50
E. B.	Prednisolone	50	—	0	Prednisolone	30

*Cortisone was administered intramuscularly as cortisone acetate. Prednisolone was given intravenously as prednisolone hemisuccinate. All other corticosteroids were administered orally.

test. Supplementary adrenal steroid therapy was given before operation to 11 patients. Table II shows the hormones and dosages employed.

Samples of venous blood were collected before and at 2, 4, and 6 hours after the beginning of all ACTH infusions. Armour ACTH was used in these studies. On the day of operation, blood was drawn prior to anesthesia and at varying intervals postoperatively in all patients. Blood was also obtained during the surgical procedure in 5 of them.

Plasma-free 17-hydroxycorticosteroids

(17-OHCS) were measured by a modification of the technique of Nelson and Samuels.[13] Normal values for plasma 17-OHCS, determined by the method described, range from 6 to 27 γ per cent with a mean of 18 ±5 γ per cent.

RESULTS

Clinical observations. A summary of pertinent clinical data on the patients selected for study may be seen in Tables I and II. In these tables are included the dosage of the various hormones received by the patients, both as maintenance therapy for the medical disease and as supplementary therapy in preparation for operation. It may be observed that fluorocortisol and triamcinolone were equally effective in protecting the patients from the stress of the operation. Their effectiveness in this regard appears to be independent of their action on renal sodium excretion, for fluorocortisol is a potent cause of sodium retention, whereas triamcinolone has been shown to produce sodium diuresis in normal subjects.[11, 19]

It is of interest that the only 2 patients in our series (E. V. and F. W.) who did not receive supplementary hormone preparation for operation were the only ones who developed complications during the immediate postoperative period. E. V. underwent a diagnostic dilation and curettage performed under cyclopropane anesthesia and inadvertently was given 2 L. of 5 per cent glucose in water over a 5-hour period postoperatively. At the completion of this infusion, the patient developed a moderate adrenal crisis, which was treated promptly with the intravenous administration of 50 mg. of prednisolone hemisuccinate. F. W. underwent a 2-hour repair of a ruptured Achilles tendon without incident. However, 3 hours postoperatively, an acute generalized exacerbation of neurodermatitis appeared. The skin lesions were treated successfully by a single injection of 50 mg. prednisolone hemisuccinate.

Hormone studies. The effect of a standard infusion of ACTH on the level of free plasma 17-OHCS was determined in 12 patients preoperatively (Table III). It is apparent that the preoperative free plasma 17-OHCS response to ACTH in 11 of these subjects was diminished in comparison with the mean values for the control group.

Table III. *Effect of ACTH infusions on free plasma 17-OHCS levels*

	Preoperative 17-OHCS levels (γ/100 ml.)			
		Hours		
*Subjects**	*Control*	*2*	*4*	*6*
J. K.	8	11	14	21
J. G.	14	20	27	28
R. L.	15	16	21	25
H. D.	18	29	32	34
P. D.	12	14	17	21
B. O.	19	30	37	42
L. F.	9	13	18	20
J. F.	11	15	21	18
H. C.	10	14	19	19
F. W.	7	6	8	10
E. V.	13	16	18	17
E. D.	7	12	13	16
Normal mean (15 subjects)	18.4	33.0	40.2	46.1
S.E.	±2.1	±3.0	±3.3	±3.6

*All patients received a 6-hour infusion of 30 I.U. of ACTH.

The free plasma 17-OHCS values in response to operation in all of the patients studied are recorded in Table IV. During the immediate postoperative period, the free plasma 17-OHCS levels reached in 10 patients were definitely less than we have observed in patients with normal adrenal function who underwent similar surgical procedures.[9, 12] In J. G., H. D., and B. O., levels of free plasma 17-OHCS developed after operation which were within our normal range for these operations.[9, 12]

Three studies are presented in detail to illustrate some of the physiologic principles involved in the hormonal management of patients with adrenal insufficiency who are to undergo operations.

Study 1. E. D., a 63-year-old man, entered the hospital with recurrent nausea and vomiting associated with a 30-pound weight loss over a 3-month period. He had been taking cortisone or prednisone continuously for 4 years and for 1 year, respectively, as therapy for rheumatoid arthritis (Table I). A gastrointestinal survey showed

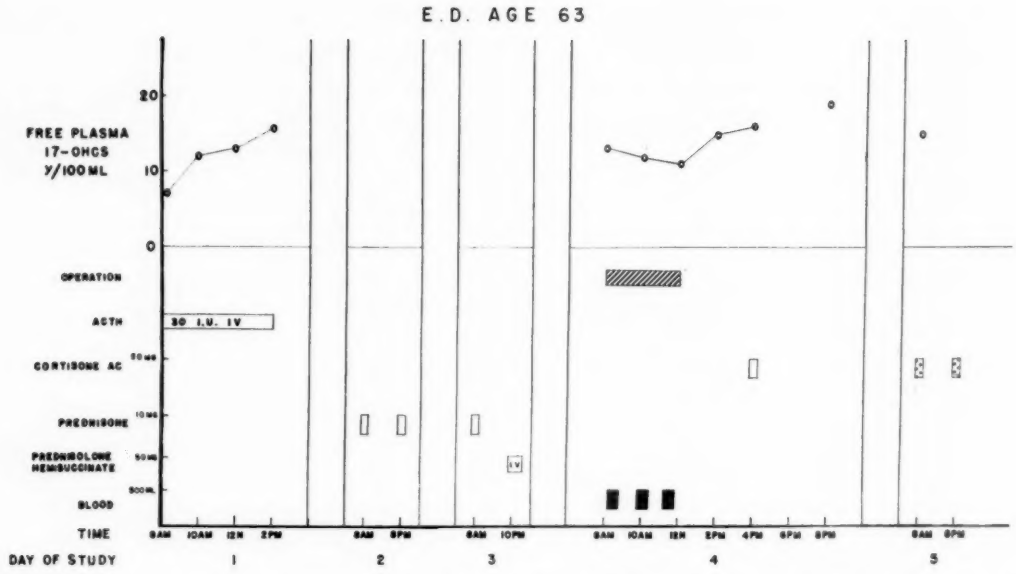

Fig. 1. Response of free plasma 17-OHCS levels to ACTH, corticosteroids, and operation in E. D. The hours and days of study are plotted along the abscissa. The dosages of hormones and the concentration of free plasma 17-OHCS are indicated along the lower and upper part of the ordinate, respectively.

an extensive carcinoma of the stomach. The patient was given a 6-hour infusion of ACTH 3 days preoperatively and was prepared for operation by a 30-minute infusion of 50 mg. of prednisolone hemisuccinate 10 hours before operation. He underwent a subtotal gastrectomy (80 per cent of the stomach was resected) and received 3 units of blood during the procedure. Blood pressure and pulse remained remarkably stable during the immediate postoperative period. A single intramuscular injection of 50 mg. of cortisone acetate was given 4 hours postoperatively as routine steroid coverage (Fig. 1).

The patient's clinical course was uneventful until the evening of the fourth postoperative day, when he complained of lower abdominal pain and suddenly went into shock. Although intravenous fluids containing cortisol and norepinephrine were administered promptly, the patient died during the early morning hours. Diffuse peritonitis secondary to a perforation of the gastroduodenal suture line was found at autopsy. Both adrenal glands were markedly atrophic and microscopically only a few strands of cortical cells could be identified within the zona fasciculata and zona reticularis.

Endocrine data. The infusion of ACTH 3 days preoperatively produced an elevation in the level of free plasma 17-OHCS from 7 γ to 16 γ per 100 ml. The administration of 50 mg. of prednisolone hemisuccinate 10 hours preoperatively resulted in a plasma 17-OHCS value of 13 γ per 100 ml. at the onset of anesthesia. Four hours postoperatively, free plasma 17-OHCS had increased to 16 γ per 100 ml. Four hours after the injection of 50 mg. of cortisone acetate, the free plasma 17-OHCS level was only 19 γ per 100 ml. (Fig. 1).

Comment. Five years of corticosteroid therapy resulted in profound adrenocortical suppression in this subject, as indicated by the minimal plasma 17-OHCS response both to ACTH and to operation and by the autopsy finding of marked adrenal cortical atrophy. Although none of the blood samples taken on the day of operation showed a free plasma 17-OHCS value above the range of normal, this patient tolerated a major surgical procedure without incident until the fourth postoperative day. His sudden death was due to an acute suppurative peritonitis associated with peripheral vascular collapse which was unresponsive to intravenously administered cortisol and norepinephrine.

Study 2. E. B., a 65-year-old man, entered the hospital with suprapubic pain and urinary frequency of 2 months' duration secondary to a vesical calculus. Ten months before admission, he had undergone a total bilateral adrenalectomy for metastatic carcinoma of the prostate with only fair clinical results. During this period he had been successfully maintained on a divided daily oral dose of 50 mg. of cortisone acetate. He was started on 2 mg. of fluorocortisol a day for 1 week before operation. At 10 P.M. on the evening before operation, 50 mg. of prednisolone hemisuccinate was administered intravenously over a 30-minute interval. The next morning a large vesical calculus was removed through a supra-

pubic incision. The patient received 1 unit of blood during the operation and was returned to the recovery room in good condition. Two hours postoperatively, he became nauseated and vomited twice. Although his vital signs remained stable, he was given a rapid intravenous injection of 30 mg. of prednisolone hemisuccinate. No further hormone therapy was administered until the next morning, at which time the patient felt extremely well. The remainder of his postoperative course was uneventful. It is of interest that this patient received 315 mEq. of sodium (as NaCl) intravenously on the day of operation and excreted 357 mEq. of sodium in the urine over the same 24-hour period.

Endocrine data. This totally adrenalectomized patient had a free plasma 17-OHCS level of 6 γ per 100 ml. at 10 P.M. on the evening before the operation. Ten hours after the infusion of 50 mg. of prednisolone hemisuccinate, the plasma 17-OHCS was 10 γ per 100 ml. This level had decreased to 6 γ per 100 ml. by the end of the operation (Fig. 2). Eight hours after the second intravenous administration of prednisolone hemisuccinate, the free plasma 17-OHCS value was 15 γ per 100 ml. and by the first postoperative day it had fallen to 6 γ per 100 ml.

Comment. This case demonstrates the efficiency of prednisolone, a potent glucocorticoid, in the protection of a totally adrenalectomized patient during a period of major surgical stress. This subject's entire postoperative course was uncomplicated except for transient nausea and vomiting 2 hours after the operation. Frequent blood samples obtained over a 24-hour period on the day of operation showed that free plasma 17-OHCS values were within our lower range of normal for at least 16 hours. Although this patient was shown to be in negative sodium balance on the day of operation, the serum sodium concentration was 142 mEq. per liter on the morning of the first postoperative day.

Study 3. This study was an endeavor to determine the duration of action of prednisolone in allowing an adrenalectomized animal to survive surgical stress. An 8 kilogram male dog was bilaterally adrenalectomized and postoperatively was maintained on 0.5 mg. of desoxycorticosterone acetate (DOCA) given intramuscularly plus a 1 Gm. sodium chloride supplement per day. Three weeks after the adrenalectomy, food, water, DOCA, and salt were withheld for 24 hours prior to operation. Then, under ether anesthesia, a 3 inch slit was made in the central abdominal wall through which small loops of intestine were successively exteriorized and gently pinched and stretched until the length of small bowel had been manipulated for 20 minutes. After recovering from anesthesia, the animal was placed back in his cage without food or water. Blood pressure readings from a femoral artery and hemoglobin and hematocrit measurements were taken at repeated intervals before and after operation (Table V). Ten hours after operation, the dog was semicomatose and in a state of peripheral vascular collapse with a mean femoral arterial blood pressure of 65 mm. Hg. The hematocrit had risen from 39 to 44 per cent. At this time 4 mg. of dexamethasone phosphate was rapidly injected intravenously. The same dose was repeated 1 hour later. The following morning (26 hours after trauma), the dog was still weak but was alert and hungry. The arterial pressure had risen to 93 mm. Hg and the hematocrit had decreased to 38 per cent. The dog was given another injection of 4 mg. dexamethasone phosphate and was allowed

Table IV. *Effect of surgical operations on free plasma 17-OHCS levels*

Subjects	*Operation*	*Free plasma 17-OHCS (γ/100 ml.)*				
		Before anesthesia	*During operation*	*Hours postoperatively*		
				0	*2*	*4*
J. K.	Subtotal colectomy	6	21	25	22	—
	Closure of colostomy	12	—	12	15	—
J. G.	Cholecystectomy	11	26	33	—	27
R. L.	Transurethral resection	11	9	19	28	25
H. D.	Subtotal colectomy	18	31	46	55	50
P. D.	Inguinal herniorrhaphy	9	—	7	18	19
B. O.	Subtotal colectomy	12	—	33	42	57
L. F.	Drainage of osteomyelitis	6	—	7	10	13
J. F.	Nailing of femur	8	—	6	9	14
H. C.	Femoral osteotomy	7	—	5	15	12
F. W.	Tenorrhaphy	4	—	4	7	—
E. V.	Dilation and curettage	11	—	10	11	14
E. D.	Subtotal gastrectomy	13	12	11	15	16
E. B.	Suprapubic cystolithotomy	10	—	6	7	—

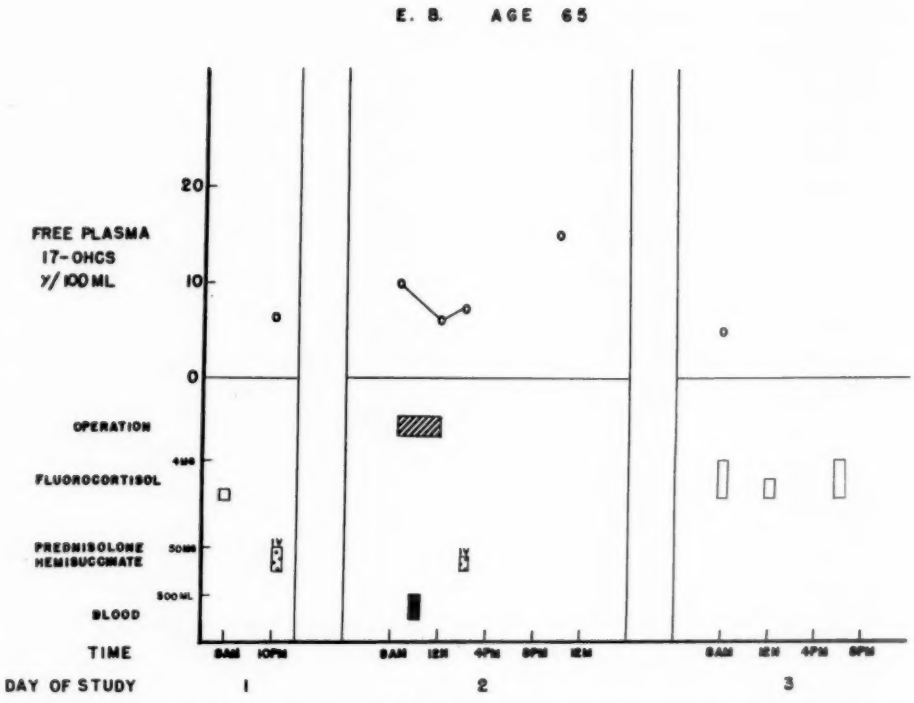

Fig. 2. Response of free plasma 17-OHCS levels to corticosteroids and operation in E. B. The data are plotted as indicated in the legend to Fig. 1.

to have food and water for the first time in over 48 hours. He was again started on 0.5 mg. DOCA and 1 Gm. of salt supplement daily.

Two weeks later, when the animal was once again in good condition, the experiment was repeated exactly as described with one important exception—the dog received a single rapid intravenous injection of 20 mg. of prednisolone hemisuccinate 24 hours before operation. The animal did not go into adrenal crisis after intestinal manipulation under ether anesthesia. Although the dog had been deprived of food and water for 48 hours, on the morning after operation he was very hungry and normally active. The arterial pressure was well maintained. However, the hematocrit had risen to 41 per cent 24 hours postoperatively (Table V).

Hormone studies. Blood samples were obtained immediately before the operation during both experiments for free plasma 17-OHCS analysis. Only a trace amount of free 17-OHCS (less than 1 γ per 100 ml.) was detectable in any of the specimens.

Comment. This experiment illustrates the extreme sensitivity of an adrenalectomized animal to surgical trauma. It also demonstrates the long duration of action of prednisolone in protecting an adrenalectomized dog through a surgical stress, although the plasma level of prednisolone had been barely detectable preoperatively.

DISCUSSION

From the data presented in this study it appears that prolonged corticosteroid therapy in man reduces the adrenal cortical capacity to respond to exogenous ACTH and to surgical trauma. These results furnish biochemical support for numerous clinical observations concerning the dangers of surgical stress in patients who have been receiving corticosteroid therapy prior to operation.[5, 16] The duration of adrenal suppression after cessation of steroid therapy is unknown. Certain patients may respond normally to exogenous ACTH within 1 or 2 days after treatment is stopped, whereas others require weeks of ACTH administration for restoration of normal adrenocortical responsiveness.[3, 15] Although 3 of our patients developed free plasma 17-OHCS levels postoperatively which were within our normal range for these operations, the majority of our patients did not demonstrate a normal plasma 17-OHCS response to their operation. Consequently, all patients who have been maintained on corticosteroid ther-

apy prior to operation should receive supplementary adrenocortical steroid hormone therapy in preparation for operation, no matter how minor the surgical procedure.

In order to select a therapeutic program that will allow a patient with corticosteroid-induced adrenal suppression to withstand a surgical operation, it is essential to be cognizant of the physiology involved in the development of an adrenal crisis postoperatively. Swingle and colleagues[17] have demonstrated that the greatly increased susceptibility of adrenalectomized dogs to stress induced by a minor operative procedure is apparently due to severe dehydration of the extracellular compartment owing to the passage of water into cells. Plasma volume and blood pressure decline, eventuating in circulatory collapse. The fluid so shifted is held immobile within the cells, presumably by osmotic forces which the animal is powerless to overcome in the absence of glucocorticoids despite the desperate need to rehydrate the extracellular space and maintain circulation. When a glucocorticoid is injected, the adrenalectomized animal is able to redistribute intracellular fluid and increase the plasma volume, thus restoring circulatory efficiency.[17] It is important to bear in mind that potent mineralocorticoids, such as aldosterone or desoxycorticosterone, exert only a minimal effect on the distribution of intracellular fluid and are incapable of relieving the severe symptoms of adrenal insufficiency, either in dogs or man.[10, 18]

It seems appropriate to consider the relationship between the plasma level of a glucocorticoid and its biologic activity. In discussing this relationship, it must be remembered that the recently developed synthetic corticosteroids are more effective physiologically on a weight basis than cortisol, a naturally occurring adrenal hormone.[8, 19] Two possibilities exist as explanations for this quantitative difference in potency between the synthetic corticosteroids and cortisol. First, the synthetic cortisol analogues, of which prednisolone is representative, may be metabolized at a slower rate by the liver than is cortisol.[14] Indeed, prednisolone has been shown to have a biologic half-life of 191 minutes in human plasma after its infusion while the corresponding half-life of cortisol is only 114 minutes.[2, 12] This observation indicates that a given dose of prednisolone should remain in the circulating blood longer before it is completely metabolized than an identical amount of cortisol. That a decreased clearance rate of prednisolone from the blood is solely responsible for its enhanced biologic activity seems unlikely. A second explanation is suggested by the work of Glenn, who injected 1.0 mg. of prednisolone and 1.0 mg. of cortisol (in 0.1 ml. of ethanol) into the tail veins of fasted adrenalectomized rats and found that the

Table V. *Blood pressure and hemoconcentration changes of an adrenalectomized dog after intestinal manipulation with and without pretreatment with prednisolone*

*Experiment No.**	*Hours after operation*	*Condition of animal*	*Blood pressure (mm. Hg)*	*Hemoglobin (Gm. %)*	*Hematocrit (%)*	*Dose steroid† (mg.)*
1	Initial	Normal	122	12.5	39	—
	8	Weak	86	13.6	42	—
	10	Shock	65	14.1	44	4
	11	Shock	71	—	—	4
	26	Weak	93	11.8	38	4
2	Initial	Normal	117	12.0	38	—
	8	Normal	120	12.2	39	—
	10	Normal	126	12.5	39	—
	24	Normal	112	12.9	41	—

*In experiment No. 1, no pretreatment with prednisolone was given. In experiment No. 2, 20 mg. of prednisolone hemisuccinate was injected intravenously 24 hours before operation.

†Dexamethasone phosphate was administered intravenously to treat the adrenal crisis which occurred postoperatively in experiment No. 1.

liver glycogen deposition response to prednisolone lasted for 32 hours, whereas cortisol exerted a similar effect for only 8 hours.[6] The results of this experiment suggest that prednisolone may persist for longer periods in tissue fluids than does cortisol. Thus, after prednisolone has completely disappeared from the blood, it still may be capable of influencing metabolic processes operating at a cellular level.

In this regard, it is pertinent to consider our experiment with the adrenalectomized dog. A single intravenous injection of 20 mg. of prednisolone hemisuccinate administered 24 hours preoperatively prevented the animal from going into adrenal crisis after intestinal manipulation under ether anesthesia. Our inability to detect a significant quantity of free plasma 17-OHCS in this dog's plasma immediately before operation may be due to the fact that the plasma half-life of prednisolone hemisuccinate is only 80 minutes in the dog.[4] It appears that prednisolone exerted a protective effect on a fasted adrenalectomized dog's capacity to withstand surgical stress at a time when it was barely detectable in the plasma.

That this observation may have clinical application in man is suggested by our study of E. D., who was shown at autopsy to have marked bilateral adrenocortical atrophy. This patient received a 30-minute infusion of 50 mg. prednisolone hemisuccinate 10 hours before subtotal gastrectomy. Although the free plasma 17-OHCS levels varied between 12 and 16 γ per 100 ml. during operation and in the immediate postoperative period, he did not develop signs of adrenal insufficiency after the surgical procedure. If one considers these plasma 17-OHCS values to be within the range of normal, using cortisol as a standard of reference, it appears that high blood levels of prednisolone are not essential during operation in order to protect a patient with adrenocortical atrophy undergoing a major operation. The potency of prednisolone in this respect may be related primarily to its persistent action in the tissues rather than to its actual blood level.

It is not within the scope of this paper to present a detailed method of how to prepare subjects with adrenocortical suppression for operation. It is evident from our data that a variety of different techniques have proved to be effective in this situation. However, we would like to suggest one clinical area in which the oral administration of steroid hormones both before and after operation may be desirable. An operation often is necessary in orthopedic patients, who have been on prolonged corticosteroid therapy, for the repair of an infected or fractured bone which may be osteoporotic. We have observed that by carrying these patients through their operation with supplementary adrenal steroids administered orally, it is easier to reduce their steroid dosage rapidly postoperatively.

SUMMARY

The free plasma 17-OHCS response to the preoperative intravenous administration of ACTH and to surgical operation was measured in 12 patients who had been receiving prolonged corticosteroid therapy. The plasma 17-OHCS response to preoperative ACTH administration was diminished in 11 patients and in 9 of them the free plasma 17-OHCS levels noted postoperatively were definitely less than we have observed in patients with normal adrenal function who underwent similar surgical procedures. The only 2 patients who did not receive supplementary hormone preparation for the operation were the only subjects who developed complications during the immediate postoperative period, even though their operative procedures were minor. It was not possible to correlate the adrenocortical response to operation with either the total dosage or the duration of corticosteroid therapy in our series of patients.

Evidence is presented which suggests that prednisolone can exert a protective effect on the ability of a patient with adrenal insufficiency to withstand a major surgical stress for many hours after free plasma 17-OHCS values have fallen to within the lower range of normal. It appears that the newer cortisol

analogues, such as prednisolone, are still capable of influencing metabolic processes operating at a cellular level after they have disappeared from the circulating blood.

The authors are grateful to Dr. C. J. Szmal, Schering Corporation; Dr. N. E. Capeci, Merck Sharp & Dohme; Dr. C. H. Demos, Lederle Laboratories; Dr. R. C. Merrill, Squibb Institute for Medical Research; and Dr. D. A. Shields, Ciba Pharmaceutical Products, Inc., for supplying the corticosteroids used in this study.

REFERENCES

1. Bunim, J. J., Black, R. L., Lutwak, L., Peterson, R. E., and Whedon, G. D.: Studies on dexamethasone, a new synthetic steroid, in rheumatoid arthritis—a preliminary report, Arthritis Rheum. **1:** 313, 1958.
2. Campbell, D. W., Marks, L. J., and Karger, R.: Effect of surgical operation on the plasma clearance of infused prednisolone, Ann. Surg. **152:** 22, 1960.
3. Christy, N. P., Wallace, E. Z., and Jailer, J. S.: Comparative effects of prednisone and of cortisone in suppressing the response of the adrenal cortex to exogenous adrenocorticotropin, J. Clin. Endocrinol. **16:** 1059, 1956.
4. Collins, E. J., Forist, A. A., and Nadolski, E. B.: Delta-1-hydrocortisone and hydrocortisone: plasma 17-hydroxycorticosteroid concentrations in the dog following oral and intravenous administration, Proc. Soc. Exper. Biol. & Med. **93:** 369, 1956.
5. Fraser, C. G., Preuss, F. S., and Bigford, W. D.: Adrenal atrophy and irreversible shock associated with cortisone therapy, J. A. M. A. **149:** 1542, 1952.
6. Glenn, E. M.: In vitro and in vivo metabolism of prednisolone: studies concerning its biological effectiveness, Endocrinology **64:** 373, 1959.
7. Jenkins, D., and Schemmel, J. E.: Metabolic effects of 6-methylprednisolone, Metabolism **7:** 416, 1958.
8. Kupperman, H. S., Ho, P., and Epstein, J. A.: Comparative pharmacologic studies on prednisone, 6-methylprednisolone, and triamcinolone, Metabolism **7:** 463, 1958.
9. LeFemine, A. A., Marks, L. J., Teter, J. G., Leftin, J. H., Leonard, M. P., and Baker, D. V., Jr.: The adrenocortical response in surgical patients, Ann. Surg. **146:** 26, 1957.
10. Lipsett, M. B., West, C. D., MacLean, J. P., and Pearson, O. H.: Adrenal function after hypophysectomy in man, J. Clin. Endocrinol. **17:** 356, 1957.
11. McGavack, T. H., Kao, K. T., Leake, D. A., Bauer, H. G., and Berger, H. E.: Clinical experiences with triamcinolone in elderly men, Am. J. M. Sc. **236:** 720, 1958.
12. Murray, J. O. S., Marks, L. J., Colombo, F. V., Josephs, B., Leftin, J. H., and Leonard, M. P.: The effect of surgical operation on the plasma clearance of infused cortisol, Ann. Surg. **148:** 951, 1958.
13. Nelson, D. H., and Samuels, L. T.: A method for the determination of 17-hydroxycorticosteroids in blood: 17-hydroxycorticosterone in the peripheral circulation, J. Clin. Endocrinol. **12:** 519, 1952.
14. Nugent, C. A., Eik-Nes, K., and Taylor, F. H.: A comparative study of the metabolism of hydrocortisone and prednisolone, J. Clin. Endocrinol. **19:** 526, 1959.
15. Sandberg, A. A., Eik-Nes, K., Migeon, C. J., and Koepf, G. F.: Plasma 17-hydroxycorticosteroids in hyperfunction, suppression, and deficiency of adrenal cortical function, J. Lab. & Clin. Med. **50:** 286, 1957.
16. Slaney, G., and Brooke, B. N.: Postoperative collapse due to adrenal insufficiency following cortisone therapy, Lancet **1:** 1167, 1957.
17. Swingle, W. W., DaVanzo, J. P., Crossfield, H. C.: Glenister, D., Osborne, M., Rowen, R., and Wagle, G.: Glucocorticoids and maintenance of blood pressure and plasma volume of adrenalectomized dogs subjected to stress, Proc. Soc. Exper. Biol. & Med. **100:** 617, 1959.
18. Swingle, W. W., DaVanzo, J. P., Crossfield, H. C., Glenister, D., Osborne, M., and Wagle, G.: Effect of various adrenal steroids on internal fluid and electrolyte shifts on fasted adrenalectomized dogs, Proc. Soc. Exper. Biol. & Med. **99:** 75, 1958.
19. Thorn, G. W., Renold, A. E., Morse, W. I., Goldbien, A., and Reddy, W. J.: Highly potent adrenal cortical steroids: structure and biologic activity, Ann. Int. Med. **43:** 979, 1955.

The effect of intraduodenal administration of hypertonic glucose solution on external pancreatic secretion

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Postgastrectomy dumping symptoms have been shown to be associated with a decrease in blood volume.[9] Volume measurements of the fluid secreted into the jejunum after the introduction of hypertonic solutions indicate a rough parallelism between the amount of fluid secreted and the "measured" loss of water from the plasma and red blood cells.[5] It is not clear, however, whether all of this secreted fluid is of intestinal origin or whether the administration of hypertonic solutions also causes a secretion of pancreatic juice.

Carbohydrates have been shown to be the most effective "trigger" for inducing the dumping syndrome and cause a greater loss of water from the circulating blood than equivalent amounts of protein or fat. Presumably this is due to a greater susceptibility of carbohydrate to enzymatic action and rapid evolution of a hypertonic solution within the jejunum. The observation that a high carbohydrate intake after gastrectomy may significantly reduce fat absorption[6, 11] also suggested the need for investigation of possible changes in pancreatic secretion during the dumping syndrome.

Our studies indicated that in unanesthetized dogs there is a marked suppression of external pancreatic secretion after the intraduodenal administration of hypertonic glucose solution.

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METHODS

Seven normal dogs were prepared with Thomas duodenal cannulas (as modified by Scott and his associates[10]). The large cannula was placed surgically into the duodenum opposite the orifice of the major pancreatic duct. At the time of laparotomy for this purpose the minor pancreatic duct was ligated at the point where it normally enters the common bile duct. At a secondary operation, a similar cannula was placed into the antrum of the stomach to allow drainage of gastric secretion and to prevent passage of acid juice into the duodenum during the experiments. Until complete recovery or when an experiment was not in progress, threaded obturators were placed in the external openings of the cannulas. Gastrointestinal function was thereby undisturbed and there were no fluid losses except during the short experimental periods. At the time of the experiments, the pancreatic juice was collected by means of a fine glass cannula which was introduced into the main pancreatic duct through the duodenal cannula itself (Fig. 1).

Because of the very low rate of pancreatic secretion in the fasting dog, stimulation of pancreatic secretion was accomplished by constant secretin infusion. A solution of 0.9 per cent saline was prepared which con-

tained 0.50 to 0.67 Ivy units of secretin* per milliliter, and this was instilled through a venous catheter by a constant infusion pump. The rate of infusion employed was in the range of 1 to 2 ml. per minute.

The experiments were carried out with the dog standing upright in a Pavlov frame. The dogs were fasted for 24 hours before the experiments and the cannulation of the pancreatic duct was limited to weekly intervals for each dog to minimize the hazard of infection. The gastric cannula was opened to allow external drainage of gastric juice during the measurements of pancreatic secretion. Although the dogs occasionally developed some degree of malnutrition, they have generally been in active good health for many months.

The general plan was similar for all experiments. After cannulation of the pancreatic duct, the constant secretin infusion was begun. The volume of pancreatic juice was measured in a calibrated test tube and was recorded at 5-minute intervals. During the control period a balloon catheter was introduced into the external duodenal cannula and threaded distally in the duodenum. The balloon was then inflated to eliminate loss of any fluids introduced into the distal duodenum during the course of the experiment.

After the pancreatic secretory rate had been stable for a period of 30 to 60 minutes, the test solution was introduced distally through the duodenal catheter. In most experiments this was 150 ml. of 50 per cent glucose solution. In some instances 8 per cent sodium chloride solution, 5 per cent dextrose solution, 0.9 per cent saline solution, or distilled water was used as the test solution. Volumetric measurements of the rate of pancreatic secretion were continued at 5-minute intervals and samples were collected for analysis of total carbon dioxide, chloride, amylase, and lipase determinations. Laboratory methods utilized for these studies have been previously reported.[4] Blood pressure was measured (in some experiments) with a simple mercury manometer.

*Secretin in these experiments was provided by James D. Hammond, M.D., of Lilly Laboratory for Clinical Research.

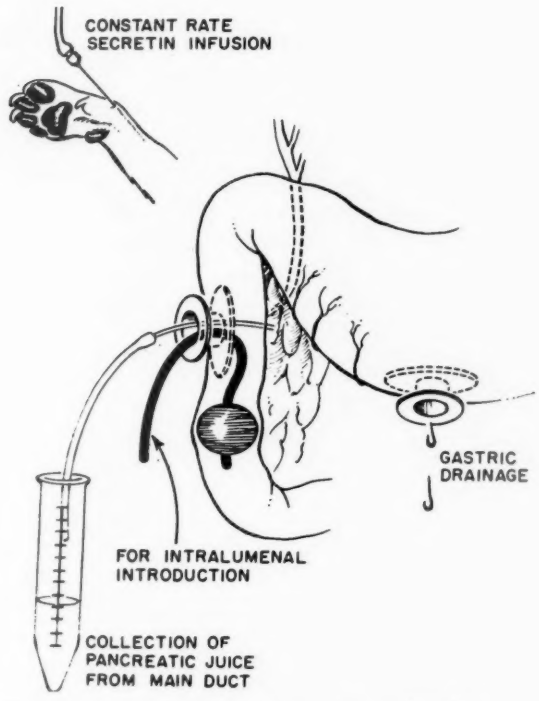


Fig. 1. Diagram of experimental preparation.

The pancreatic response after a glucose test meal was studied in similar experiments that were modified by the intravenous administration of hexamethonium bromide or replacement infusions of normal saline solution, dextran, or whole blood.

Three of the 7 dogs studied underwent a right thoracotomy with supradiaphragmatic section of both vagus nerves after the control studies. They were subjected to additional pancreatic studies after recovery from the operative procedures. Two of these dogs were also subjected to bilateral total adrenalectomy and, after this procedure, they were maintained with cortisone. These dogs were also subjected to repeat studies of pancreatic secretion after satisfactory recovery from adrenalectomy.

RESULTS

Volume of pancreatic secretion.

Variation in control secretory rate. By constant infusion of secretin solution, flow

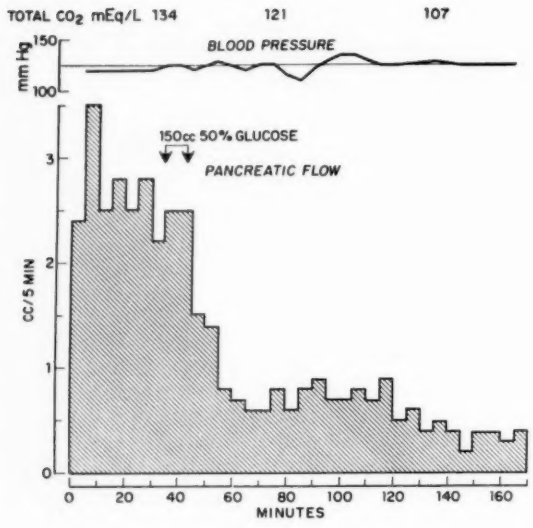


Fig. 2. Typical experiment with change in pancreatic secretion after intraduodenal hypertonic glucose administration (constant secretin infusion).

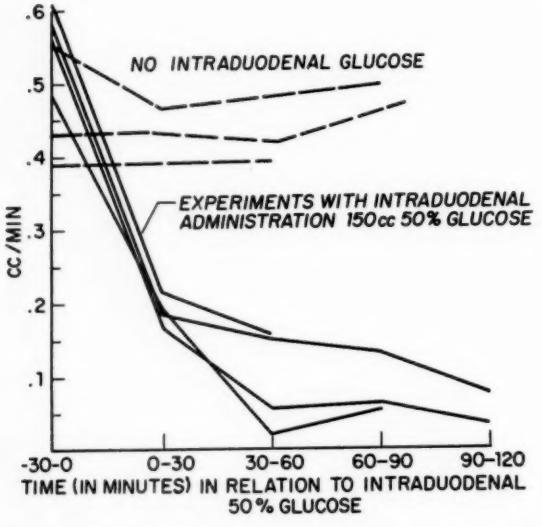


Fig. 3. Pancreatic response to intraduodenal hypertonic glucose administration compared with control studies (constant secretin infusion).

rates of pancreatic juice were maintained in the range of 8 to 29.7 ml. for 30-minute control periods (0.26 to 0.99 ml. per minute). This is approximately 10 times the secretory rate in fasted, unstimulated dogs. In 7 experiments, control secretory rates were measured for longer periods which ranged from 1 to 4 hours without other experimental manipulation. When the volume output for 30-minute periods was compared during these constant secretin infusion experiments, the greatest alteration noted was a 14 per cent shift from the mean flow rates. If 5-minute periods were compared, the alteration from period to period was often greater than this, but in many experiments the secretory rate for individual 5-minute time intervals was amazingly constant. There was no "fatigue" mechanism in regard to volume output noted over the time intervals utilized in these experiments.

Pancreatic response to intraduodenal administration of glucose (50 per cent). In 16 experiments, 150 ml. of 50 per cent glucose was instilled into the duodenum and a marked suppression of pancreatic secretion occurred despite the continuous secretin infusion. There was a "lag" period of approximately 5 minutes before the decrease in pancreatic flow rate became apparent. The change in volume of pancreatic juice in a typical experiment is depicted in Fig. 2, which also demonstrates the minimal changes in blood pressure that occurred. For individual experiments, the volume of pancreatic juice during the 30-minute period *after* glucose administration was 8.5 to 48.3 per cent of the volume obtained during the control period (Table I). In these 16 experiments the mean value was 28.5 per cent of the control volume (first 30 minutes). During the second 30-minute period, in the 8 experiments in which nothing else was done that could have possibly altered secretion of pancreatic juice, the mean value for pancreatic juice output was 24.3 per cent. There was minimal improvement in pancreatic flow in these experiments during a third 30-minute period. The mean flows for these 30-minute periods in 4 long-term experiments are shown graphically in Fig. 3.

A similar study was carried out with the low flow rates that are observed in the fasting animal not receiving secretin. The control flow was 0.068 ml. per minute over a 2-hour period but there was considerable fluctuation in this minimal flow range. After 150 ml. of 50 per cent glucose solution had been introduced into the duodenum, there

was no reduction in flow for the first 15 minutes (0.073 ml. per minute), but the reduction in flow rate over the next two 30-minute periods was striking (0.013 and 0.007 ml. per minute).

Pancreatic response to intraduodenal administration of hypertonic saline solution. Two dogs were subjected to experiments which were identical to those described except for the substitution of 8 per cent saline solution for hypertonic glucose solution. Suppression of pancreatic secretion occurred in a manner similar to that observed with hypertonic glucose. The values for the 30-minute period after introduction of the test solution were 31.6 and 15.2 per cent of the control secretory rates (Table I).

Pancreatic response to intraduodenal administration of isotonic solutions. One dog that had demonstrated marked suppression of pancreatic secretion with intraduodenal hypertonic glucose solution was then subjected to the same volume of 0.9 per cent sodium chloride solution on one occasion and 5 per cent dextrose in water on another. In neither case was there a significant alteration in the rate of pancreatic flow (Table I).

Response in pancreatic volume flow to intraduodenal air instillation or administration of distilled water. Two dogs which had previously demonstrated suppression of pancreatic flow after hypertonic glucose instillation were subjected to the intraduodenal instillation of air under pressure. This resulted in a slight decrease in pancreatic flow during the subsequent 15- to 20-minute period, but this was not nearly as marked as the suppression noted with the 50 per cent glucose solution. On the graph of one of these dogs (Fig. 4), it can be seen that this alteration was very transient. After 20 minutes, control secretory rates were again achieved. Later in the same experiments, 500 ml. of distilled water was rapidly injected into the duodenum through the balloon catheter. There was no significant alteration in volume output of the pancreas of either dog after this procedure (Fig. 4).

Effect of vagus section on pancreatic response to administration of hypertonic glucose solution. The 3 dogs subjected to vagus

Table I. *Response of pancreatic secretion to various intraduodenal test meals during constant secretin stimulation*

Date	*Dog No.*	*Output of pancreatic juice in control period (ml./min.)*	*Test meal*	*Pancreatic juice output after feeding (first 30 min.)*	
				Rate (ml./min.)	*% of control*
March 10	918	0.560	50% glucose	0.170	30.4
March 23	918	0.620		0.063	10.0
March 18	913	0.373		0.180	48.3
April 9	913	0.483		0.190	39.3
April 3	653	0.387		0.033	8.5
April 16	653	0.653		0.276	42.3
April 23	653	0.993		0.200	20.1
May 7	653	0.613		0.057	9.3
May 20	653	0.610		0.213	34.9
May 5	891	0.670		0.210	31.3
May 14	891	0.757		0.173	22.9
May 27	891	0.580		0.187	34.4
April 7	820	0.600		0.140	23.3
May 5	777	0.310		0.100	32.2
May 12	777	0.480		0.110	22.9
May 19	27	0.630		0.290	46.0
March 25	913	0.263	8% NaCl	0.083	31.6
March 30	918	0.660		0.100	15.2
June 18	913	0.337	5% glucose	0.367	109.0
July 10	913	0.830	0.9% NaCl	0.850	102.0

Table II. *The pancreatic response to intraduodenal administration of 50 per cent glucose (150 ml.) before and after supradiaphragmatic vagotomy*

Dog	Date	Control flow (ml./min.)	Pancreatic flow after intraduodenal glucose administration					
			0-30 min.		30-60 min.		60-90 min.	
			Rate (ml./min.)	% of control	Rate (ml./min.)	% of control	Rate (ml./min.)	% of control
891 (before vagotomy)	May 5	0.670	0.210	31	—	—	—	—
	May 14	0.757	0.173	23	—	—	—	—
	May 27	0.580	0.187	34	0.150	28	0.130	24
Mean				29		28		24
891 (after vagotomy)	June 17	1.040	0.647	62	0.383	37	0.220	21
	June 26	0.608	0.196	32	0.423	70	0.250	41
	Sept. 10	0.717	0.293	41	0.400	56	—	—
Mean				45		54		31
653 (before vagotomy)	April 3	0.387	0.033	8	—	—	—	—
	April 6	0.653	0.276	42	—	—	—	—
	April 23	0.993	0.200	20	—	—	—	—
	May 7	0.613	0.057	9	—	—	—	—
	May 20	0.610	0.213	35	—	—	—	—
Mean				23				
653 (after vagotomy)	June 24	0.493	0.220	45	—	—	—	—
777 (before vagotomy)	May 5	0.310	0.100	32	0.090	29	0.121	39
	May 12	0.480	0.110	23	0.200	42	—	—
Mean				26		35		
777 (after vagotomy)	May 19	0.640	0.210	33	0.240	38	—	—

Table III. *Pancreatic response to intraduodenal administration of 50 per cent glucose solution after bilateral adrenalectomy (2 dogs)*

Date	Control flow (ml./min.)	Pancreatic flow after intraduodenal glucose administration					
		0-30 min.		30-60 min.		60-90 min.	
		Rate (ml./min.)	% of control	Rate (ml./min.)	% of control	Rate (ml./min.)	% of control
June 18	0.483	0.190	39.3	0.020	4.1	0.050	10.3
Aug. 15	Bilateral adrenalectomy						
Sept. 2	0.523	0.100	19.1	0.013	2.5		
Sept. 9	0.592	0.133	22.5	0.113	19.1	0.057	9.6
May 19	0.630	0.420	66.7	0.290	46.0	0.210	33.3
May 21	Bilateral adrenalectomy						
June 2	0.600	0.400	66.7	0.290	48.3	0.190	31.7
June 9	0.600	0.096	16.0	0.170	28.3	0.260	43.3

section and subsequent challenge with hypertonic glucose solution (intraduodenal) also demonstrated a suppression of pancreatic secretion similar to but somewhat less pronounced than that observed prior to vagotomy (Table II).

Pancreatic response to intraduodenal administration of hypertonic glucose after bilateral total adrenalectomy. Two dogs which had previously demonstrated the typical pancreatic response to hypertonic glucose administration were subjected to the same experiment after adequate recovery from bilateral total adrenalectomy. The suppression of pancreatic flow during the first 30 minutes after administration of hypertonic glucose was indistinguishable from the response noted prior to adrenalectomy. Continued observation during 4 experiments after adrenalectomy showed continued suppression of pancreatic secretion in the subsequent 30-minute periods. There was

no question that the pancreatic response to hypertonic solution was unhampered by adrenalectomy (Table III).

Effect of replacement infusion with normal saline solution, dextran, or blood on the pancreatic response to intraduodenal administration of hypertonic glucose. There were 4 experiments in which dextran (300 to 400 ml.) was given 30 minutes after the 150 ml. of hypertonic glucose had been instilled into the duodenum. The continued suppression of pancreatic flow routinely noted in the control experiments was partially averted by this therapy as can be seen in Fig. 5. Rapid infusion of 500 ml. of normal saline solution in 2 experiments caused a similar response, but did not prevent suppression of pancreatic flow. Whole blood (250 ml.) was given to 2 dogs after the hypertonic test meal and the decrease in pancreatic flow was only partially averted. It can be seen in Table IV and Fig. 5 that the pancreatic response to the intraduodenal administration of hypertonic glucose solution was modified, but was not completely prevented, by the intravenous replacement therapy.

Pancreatic response to intraduodenal administration of hypertonic glucose during ganglionic block with hexamethonium bromide (C_6). Two of the dogs were given 2.5 to 5.0 mg. hexamethonium bromide intravenously at 5-minute intervals before and after the intraduodenal administration of hypertonic glucose. There was marked and constant dilatation of the pupils during these experiments. Blood pressure decreased 10 to 20 mm. Hg during the period of drug administration prior to glucose feeding. Minimal additional decrease in blood pressure occurred after the meal of hypertonic glucose. There was suppression of pancreatic secretion after intraduodenal administration of glucose in both experiments, and in one it was indistinguishable from the response observed in control studies (Fig. 6). In the other experiment, the suppression of volume flow was definitely less than that observed in any of the control studies but there had been a definite decrease in pancreatic flow after administration of the ganglionic blocking agent only (Fig. 7).

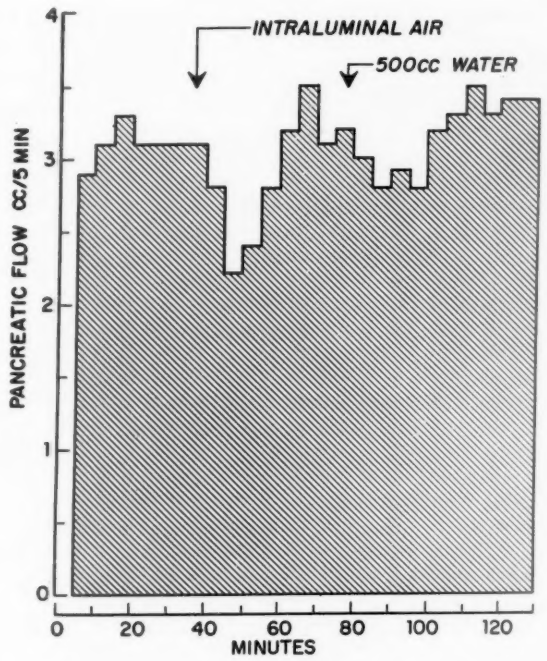


Fig. 4. Experiment demonstrating minimal change in pancreatic secretion after intraduodenal instillation of air or water.

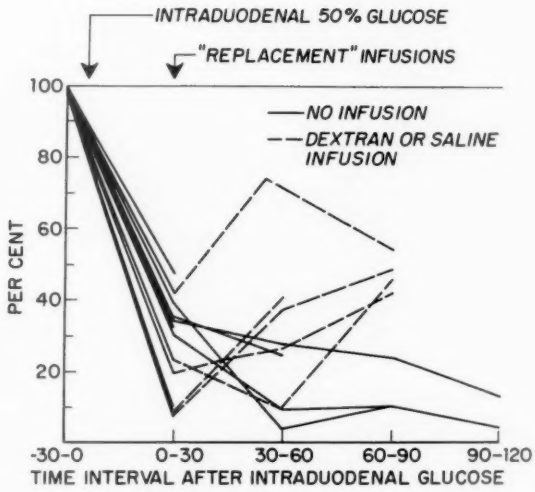
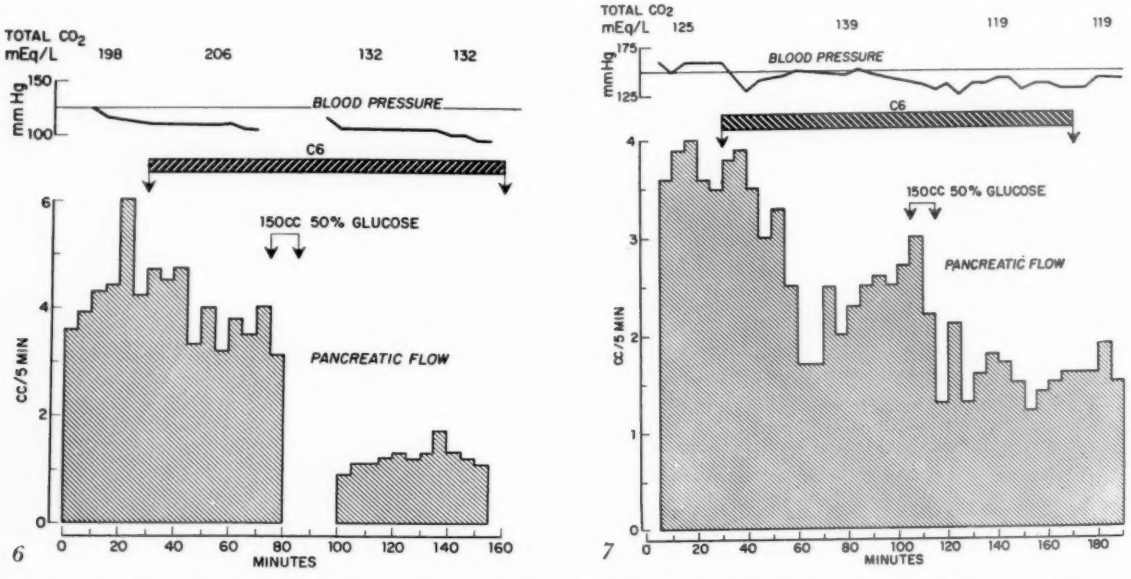


Fig. 5. The effect of intravenous replacement therapy on pancreatic suppression after intraduodenal hypertonic glucose administration.

Two dogs were subjected to the experiment described after recovery from transthoracic vagotomy. The ganglionic blocking agent was given in the same manner. The suppression of pancreatic secretion was

Table IV. *The effect of intravenous replacement therapy on the pancreatic response to intraduodenal 50 per cent glucose administration*

Dog No.	Control flow (ml./min.)	Flow after intraduodenal glucose administration		Infusion	Flow after infusion			
					First 30 min.		Second 30 min.	
		Rate (ml./min.)	% of control		Rate (ml./min.)	% of control	Rate (ml./min.)	% of control
653	0.993	0.200	20.1	Dextran (300-400 ml.)	0.265	26.7	0.420	42.3
653	0.613	0.057	9.3		0.230	37.5	0.297	48.5
913	0.320	0.210 (10″)	65.6		0.272	85.5	0.373	117.0
891	0.757	0.173	22.9		0.073	9.6	0.350	46.0
653	0.653	0.276	42.3		0.484	74.1	0.352	53.9
653	0.390	0.033	8.5	0.9% NaCl (500 ml.)	0.157	40.3	—	—
					—	—	0.320	82.1
891	0.670	0.210	31.3	Blood (250 ml.)	0.303	45.2	—	—
918	0.560	0.170	30.4	No intravenous	0.053	9.5	0.057	10.2
913	0.483	0.190	39.3	solutions	0.020	4.1	0.050	10.3
653	0.610	0.213	34.9	(controls)	0.153	25.1	—	—
891	0.580	0.187	34.4		0.150	27.6	0.130	23.9



Figs. 6 and 7. Intraduodenal administration of 150 ml. 50 per cent glucose solution to dog receiving intravenous hexamethonium bromide.

somewhat delayed in 1 experiment but the results of 2 additional experiments were identical to those already described.

Nature of pancreatic secretion.

Bicarbonate concentration. The bicarbonate concentration in pancreatic juice was determined before and after intraduodenal administration of glucose (50 per cent) in 10 of the standard experiments. The mean value for the control period was 134 mEq. per liter and for the 30 minutes after glucose administration the mean bicarbonate concentration was 118 mEq. per liter. There was a decrease in concentration at the time of decrease in rate of flow in all 10 experiments, and this ranged from 2 to 36 mEq. per liter. There were similar changes in bicarbonate concentration commensurate

with flow changes in experiments modified by vagotomy, adrenalectomy, and hexamethonium bromide administration. When a subsequent increase in flow followed an infusion of dextran or saline solution, the bicarbonate concentration increased toward the control level. There were reciprocal changes in chloride concentration with these measured alterations in bicarbonate level.

Enzyme determinations. Determinations were made only of amylase and lipase concentrations. It would appear from the data shown in Fig. 8 that, although we observed a major change in rate of secretion after intraduodenal glucose administration, there was minimal change in enzyme composition of the pancreatic juice. This was also true in experiments involving various modifications of the standard experiment.

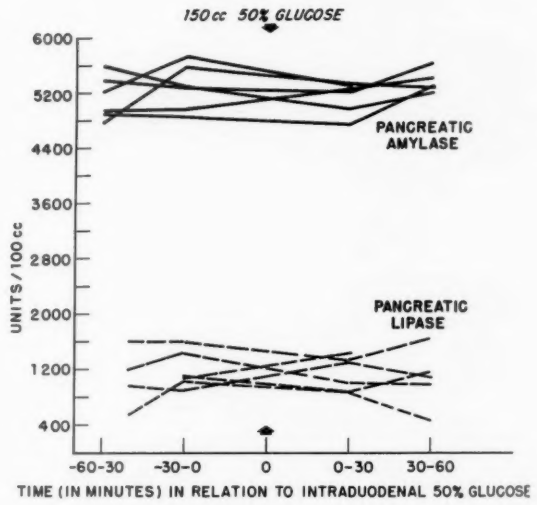


Fig. 8. Pancreatic enzyme concentration in 6 experiments on 3 dogs before and after intraduodenal administration of 150 ml. of hypertonic glucose.

DISCUSSION

The preparation utilized for these studies was very satisfactory for short-term studies of pancreatic secretion. The intermittent cannulation produced no changes in the gland itself and electrolyte replacement was not a problem as it is with permanent fistulas. The gastric cannula was probably not absolutely necessary, as adequate fasting prior to such experimental studies should suffice. The possibility of unsuspected acid evacuation into the duodenum and possible confusion resulting from endogenous secretin production led us to add this cannula to our preparation. Constant secretin infusion produced a sufficient volume flow of pancreatic juice to make changes in rate of flow very apparent.

The major change in pancreatic secretion after a hypertonic test meal of glucose was a marked reduction in rate of flow. The hypertonicity of the test meal was a significant factor since the findings were reproduced with a saline solution of similar osmolarity. Isotonic solutions of glucose or sodium chloride produced no change in secretion as has been previously reported.[2] An increase in pancreatic secretion has been described when hypotonic solutions were introduced into the bowel. In 2 experiments in this study, however, a large volume of distilled water produced no real change in rate of pancreatic secretion. These data and the lack of significant response to air distention of the intestine support the conclusion that the changes in pancreatic function after test meals were produced by the hypertonicity of the test material and not by the bowel distention that follows administration of hypertonic solutions.

In studies of the dumping syndrome it has been shown that shifts of fluid occur into the bowel in response to ingestion of hypertonic foodstuffs. The possibility that this dilution fluid originated partially from the pancreas has been effectively eliminated by these studies. Pancreatic output actually diminishes during the symptomatic "fluid loss" phase of the dumping syndrome.

A decrease in plasma volume, as well as even larger changes in effective circulatory volume, seem to accompany the dumping syndrome.[8] Actual shock was not produced in these studies as shown by the blood pressure recordings made during several of these experiments. On the assumption that the hypovolemia from these temporary losses might be the cause of pancreatic suppression, intravenous replacement with saline, plasma expander, and blood was attempted

after the suppression of pancreatic output had become apparent. Although the decrease in pancreatic output was less striking during replacement infusions, none of the solutions could acutely return the pancreatic flow rates to normal levels. When the vagus innervation of the pancreas was removed by transthoracic vagotomy, the suppression of pancreatic secretion after the hypertonic test meal was sometimes less striking. This is not surprising in view of the role the vagus nerves play in the normal control of pancreatic secretion. It seems important, however, to stress that suppression of pancreatic output did occur after the hypertonic meal despite the loss of vagus innervation. The same observations were made on normal and vagotomized dogs during administration of hexamethonium bromide, a ganglionic blocking agent that is particularly efficient for blocking the sympathetic system.

Studies by Edmonds,[3] Mann and McLacklin,[7] and Babkin,[1] have demonstrated profound suppression of external pancreatic secretion after administration of epinephrine or ephedrine. This has occurred with doses too small to elevate the blood pressure. The reduction in secretion is thought to be due to a markedly reduced blood flow through the pancreas. In view of the circulatory changes noted during the dumping episode, epinephrine appeared to be a possible explanation for the findings we have observed. However, when our experiments were repeated in dogs which had been subjected to total adrenalectomy, the findings in regard to pancreatic response were the same as in control studies. This does not eliminate the possibility that a vasopressor substance of extra-adrenal origin may be acting in a manner similar to the known effect of epinephrine on the pancreas.

The concentration of bicarbonate decreases with the reduction in flow of pancreatic juice. There is always a direct relationship between the bicarbonate concentration and volume at slow rates of flow (0.05 ml. per minute), but the decrease in bicarbonate concentration is less striking with the artificial elevations of pancreatic output.

Although enzyme determinations have a wide range of error, it appears that there are minimal changes in enzyme concentration despite the marked changes in flow rates. The total quantity of enzyme is significantly reduced, however, by virtue of the volume changes.

SUMMARY

Although we have not established the exact mechanisms involved, these studies do demonstrate a marked suppression of external pancreatic secretion in dogs during the experimental "dumping test." This may be a cumulative effect of several factors but experimental modifications such as vagus section, autonomic block, adrenalectomy, or replacement infusions did not prevent this response. The reduction in pancreatic secretion may well be a reflection of vasoconstriction and reduced blood flow in the pancreas itself. The resultant diminution in total enzyme secretion may also be partially responsible for the absorptive defects noted after total gastrectomy in man. The physiologic aspects of this changes are worthy of more intensive study for they may have direct bearing on other symptoms and circulatory findings associated with the dumping syndrome.

We wish to thank F. Washington and J. O'Flaherty for their technical assistance.

REFERENCES

1. Babkin, B. P.: Influence of the blood supply on pancreatic secretion, J. Physiol. **59:** 153-163, 1924.
2. Crider, J. O., and Thomas, J. E.: Difference in effect of distilled water and isotonic solutions in intestine on pancreatic secretion, Proc. Soc. Exper. Biol. & Med. **44:** 299-301, 1940.
3. Edmunds, C. W.: The antagonism of the adrenal glands against the pancreas, J. Pharmacol. & Exper. Therap. **1:** 135-150, 1910.
4. Hudock, J., and Lawrence, W., Jr.: Gastric control of pancreatic secretion: an experimental study, SURGERY **46:** 682-687, 1959.
5. Lawrence, W., Jr., and Mathews, D. H.: A

study of water "shift" in experimental dumping syndrome, S. Forum **10:** 180, 1960.

6. Lâwrence, W., Jr., Vanamee, P., Peterson, Ann S., McNeer, G., Levin, S., and Randall, H. T.: Alterations in fat and nitrogen metabolism after total and subtotal gastrectomy, Surg. Gynec. & Obst. **110:** 601-616, 1960.
7. Mann, F. C., and McLacklin: Action of adrenalin in inhibiting the flow of pancreatic secretion, J. Pharmacol. & Exper. Therap. **10:** 251-259, 1917.
8. Mathews, D. H., Lawrence, W., Jr., Poppell, J. W., Vanamee, P., and Randall, H. T.: Change in effective circulating volume during experimental "dumping syndrome," SURGERY **48:** 185, 1960.
9. Roberts, K. E., Randall, H. T., Farr, H. W., Kidwell, A. P., McNeer, G. P., and Pack, G. T.: Cardiovascular and blood volume alterations resulting from intrajejunal administration of hypertonic solutions to gastrectomized patients: the relationship of these changes to the dumping syndrome, Ann. Surg. **140:** 631-640, 1954.
10. Scott, V. B., Collignon, U. J., Eugel, H. J., and Johnson, O. C.: The relation of external pancreatic secretion to variations in blood sugar, Am. J. Physiol. **134:** 208, 1941.
11. Vanamee, P., Lawrence, W., Jr., Levin, S., Peterson, A. S., and Randall, H. T.: Further observations on postgastrectomy steatorrhea: the effect of high carbohydrate intake and of hydrochloric acid administration on fat absorption, Ann. Surg. **150:** 517-528, 1959.

Diverticulitis and its surgical management

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Of the many diseases which affect the colon none has been the subject of more diverse changes in the concepts of management during the past 2 decades than diverticulitis. About 20 years ago Babcock and a few others publicly favored early resection for diverticulitis of the colon rather than palliative or delayed operation. Since that time it has become generally accepted that preventive surgical intervention is a logical and justifiable procedure and that early surgical intervention for the complications of diverticulitis is mandatory to prevent prolonged morbidity and invalidism. Advances in surgical technique, preoperative and postoperative care, anesthesia, and ancillary factors in the care of patients have led to a more liberal attitude in the management of diverticulitis and it is becoming more apparent to all concerned that diverticulitis is a surgical disease from its very inception. The policy of waiting until complications occur before operation is performed is outmoded and archaic.

The purpose of this presentation is to record our experience with diverticulitis and to illustrate the rationale for early surgical intervention in uncomplicated diverticulitis as well as for the mandatory surgical treatment of complications.

MATERIAL

The material for this clinical study of diverticulitis was obtained from records of a series of patients treated in the Department of Proctology of Temple University Medical Center during a period of 19½ years from September, 1940, to May, 1960, inclusive. During this time 253 patients with diverticulitis of the colon were treated. There were 114 men and 139 women. The ages ranged from 8 years to over 85. While the majority of cases of diverticulitis occur during the sixth and seventh decades it is not unusual for the disease to manifest itself in patients who are in their twenties and thirties (Table I).

SURGICAL TREATMENT

Surgical operations have been performed on 143 of the 253 patients with diverticulitis of the colon, representing an over-all surgical incidence of 55.9 per cent. It is interesting to note the increment of operation as it increased yearly. In 1948, 30.8 per cent of the patients were treated surgically, in 1950 the incidence had risen to 35.1 per cent, and by 1955 it was 46 per cent. Altogether, since 1948 73.6 per cent have been treated surgically, representing 110 of 149 patients treated for diverticulitis since that year.

Emergency operations have been performed in 26 cases (Table II). Two deaths resulted, a mortality rate of 7.7 per cent. One patient developed a severe thrombophlebitis after drainage of a pelvic abscess and did not return for further surgical treatment. The remaining 23 patients had subsequent operations and are considered to have had definitive operations.

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Table I. *Distribution of patients according to age: authors' cases*

Age	*No. of patients*
1-20	1
21-30	5
31-40	16
41-50	22
51-60	88
61-70	84
71-80	30
81-90	7
Total	253

Five patients with diverticulitis underwent exploratory celiotomy without additional procedures. Carcinoma of the pancreas was found in one, carcinoma of the sigmoid colon with peritoneal carcinomatosis in the second, carcinoma of the liver in the third, and carcinoma of the rectosigmoid with hepatic metastases in the fourth. The fifth was a frail elderly patient who complained of recurrent episodes of left lower quadrant abdominal pain. On exploration the sigmoid was found to be bound with adhesions and since the diverticular process did not appear to be severe the adhesions were lysed and the abdomen closed.

Primary resection of the diseased portion of the colon has been a very satisfactory procedure in the majority of instances. The advantages of primary one-stage resection without concomitant colostomy are many, but proper surgical judgment must be exercised or the results may be disastrous. One-stage resections were performed in 96 patients, or 71.4 per cent of the total 135. Multiple-stage operations were carried out in 39 patients. In this group were 3 patients with small bowel fistulas and 1 with a sigmoidovesical fistula for which resections were instituted. Two deaths from peritonitis occurred in the entire group of 135 patients, which was a mortality rate of 1.5 per cent (Table III).

COMPLICATIONS

In cases of chronic diverticulitis the most frequent and important complications are stricture of the lumen of the colon and fistula formation. Stricture is the result of prolonged, low-grade, recurrent inflammatory processes which lead to extensive peridiverticulitis and pericolitis, fibrosis, and scarring. Fistula or sinus formation follows perforation of an inflamed diverticulum with abscess formation. Localized abscesses may become walled off or may extend to adjacent viscera, abdominal wall, or peritoneal cavity with a resulting fistulous tract.

In most cases diverticulitis with a radiographic diagnosis of stricture formation but without obstruction can be resected satisfactorily in one stage without colostomy. If the stricture has progressed to the point of producing total or near-total obstruction of the colon, it has been our policy to perform either a preliminary or a concomitant transverse colostomy. In many instances the obstruction has an acute onset and an emergency transverse colostomy must be performed. Frequently, in this stage of diverticulitis, it is impossible to distinguish between diverticulitis and carcinoma. While there is no relationship between carcinoma and diverticular disease this coincidental occurrence is sufficiently frequent to warrant resection as early as possible after the diagnosis of stricture is made. In our series 12 patients were admitted to the hospital with

Table II. *Emergency operations in diverticulitis*

Operation	*No. of patients*	*Deaths*	*Mortality rate (%)*
Transverse colostomy for obstruction	12	1	8.3
Transverse colostomy and drainage of abscess or perforation	9	0	—
Incision and drainage of pelvic or lumbar abscess (without colostomy)	5	1	20
Totals	26	2	7.7

Table III. *Definitive operations for diverticulitis*

Type	*Total procedures (May, 1960)*	*No. of patients*	*Deaths*	*Mortality rate (%)*
Primary resection of colon (primary resection with partial cystectomy in 10)	96	96	2	2.1
Colostomy (resection, partial cystectomy, hysterectomy in 2, closure colostomy)	30	10	–	
Colostomy (resection, hysterectomy, closure colostomy)	3	1	–	
Preliminary procedure and resection	36	18	0	0
Resection of colon with cutaneous sinus	10	3	0	0
Resection (Bloch-Mikulicz)	21	7	0	0
Totals	196	135	2	1.5

acute colon obstruction and transverse colostomy was performed in all. There was 1 operative death, a mortality rate of 8.3 per cent. The remaining 11 patients underwent subsequent resection.

Fistula formation frequently presents a difficult problem in surgical management and the selection of the proper operation for each patient is extremely important. In cases of chronic fistula between the colon and the small intestine or the uterus, without acute inflammation or suppuration, it has been our practice to excise the fistula and perform a colon resection and small bowel resection or hysterectomy in one stage without colostomy.

Cutaneous sinuses have been encountered in 3 cases. Two of these were tracts from the colon to the lumbar region and one to the abdominal wall. In all 3 cases operations were performed in 3 stages. Case 1 is illustrative of these cases.

Case 1. A 62-year-old man had undergone a transverse colostomy in March, 1956, for acute diverticulitis. An abscess developed with subsequent fistula formation from the colon to the abdominal wall near the left ilium. When the patient was seen in December, 1957, a roentgenogram of the fistula showed the tract to run cephalad from the left iliac area entering the abdominal cavity. Sigmoidectomy and fistulectomy were performed in January, 1958. After opaque visualization of the anastomosis the colostomy was closed. The patient was discharged from the hospital 10 days later after an uneventful recovery. He has remained well since that time.

Sigmoidovesicle fistulas present a different surgical problem. Early in our experience it was our custom to perform a transverse colostomy as the initial stage, a segmental resection of the sigmoid colon with partial cystectomy and suprapubic cystostomy as the second stage, and closure of the colostomy as the third stage. More recently, suprapubic cystostomy has been omitted, although an indwelling catheter is left in place for a minimum of 14 days. In this series there have been 21 patients with sigmoidovesicle (18) and/or sigmoidouterine fistulas. There were 17 men and 4 women. Of the latter, a hysterectomy had been performed in 1 and 2 had sigmoidouterovesicle fistulas. In 10 patients resection of the colon and partial cystectomy were performed in one stage without colostomy and 11 patients underwent three-stage procedures. While there were no deaths in either group, the postoperative complications with the single-stage procedure were significantly more marked than with the multiple-stage group and for this reason we consider the latter method to be preferable.

Case 2. A 44-year-old man noted the passage of gas during urination in November, 1951. Examination showed a sigmoidovesicle fistula for which a transverse colostomy and suprapubic cystostomy were performed. At consultation in January, 1952, a fistula was demonstrated on the posterosuperior wall of the bladder. Sigmoid resection including the fistula and partial cystectomy were performed on Jan. 17, 1952. On January 28 the anastomosis was visualized radiographically and the colostomy was closed. The patient was discharged on Feb. 7, 1952, after an uneventful convalescence. He has remained well.

In the operation for diverticulitis there are technical factors which we have found to be of assistance. In many instances the scarring and chronic inflammatory reaction produces a situation in which extensive abdominopelvic dissection may be extremely difficult and hazardous. To avoid damage to the ureters during dissection, ureteral catheters are introduced prior to operation. The reassurance of being able to identify the ureters by palpation and the resulting negligible incidence of ureteral damage has more than justified this procedure. While suprapubic cystostomy at the time of partial cystectomy has been omitted recently, the use of an indwelling Foley catheter for a period of 14 days is advantageous. The catheter should be checked frequently and irrigated as necessary.

When preliminary or concomitant colostomy is necessary complete fecal diversion should be obtained. This may be achieved satisfactorily with a transverse colostomy in the right upper quadrant of the abdomen. Cecostomy is unsatisfactory for fecal diversion. When bowel infection is present daily instillations of antibiotic solutions into the distal loop of the colostomy are employed. After resection of the diseased portion of the colon the transverse colostomy is not closed until the anastomotic site has been visualized radiographically. Usually on about the tenth day after resection a thin suspension of the opaque mixture is introduced into the distal loop of the colostomy to insure the patency of the lumen and the security of the anastomosis.

REHABILITATION

Every surgeon is concerned with the return of his patient to his job and useful activity after surgical therapy. The true evaluation of any surgical procedure or method of management can be based only upon the rate of rehabilitation of the patients subjected to such therapy. In this series of 135 resections there were 2 hospital deaths. Two patients have died with concomitant carcinoma and 10 are lost to follow-up. Three patients complain of periodic abdominal distress. Nevertheless, they are performing their occupational duties. One patient is convalescing after a secondary resection. Besides the 2 patients who died during operation, those who died of carcinoma (2 patients), those lost to follow-up (10 patients), and the patient who is convalescing, there remain 117 patients who are well and completely rehabilitated. This represents a rehabilitation rate of 97.5 per cent (Table IV).

Many factors are important in the rehabilitation of patients with diverticulitis. The advent of antibiotics has played a role in decreasing the extent of the inflammatory reactions. This in turn has effected an increase in the number of patients selected for one-stage operative procedures. There is a definite trend toward the avoidance of repetitive attacks, in a large measure because of the primary care given by our medical colleagues and their subsequent sound advice urging early or prophylactic resection.

SUMMARY AND CONCLUSIONS

A consecutive series of 253 patients with diverticulitis of the colon is reviewed. There were 114 men and 139 women. The average age was 67 years and the greatest incidence was in patients in their fifth and sixth decades.

A total of 143 patients were operated upon in one or more stages. Primary resection and anastomosis without colostomy was performed in 96 patients or 71.4 per cent of a total of 135 resections. Two operative

Table IV. *Diverticulitis—rehabilitation (September, 1940 to May, 1960)*

No. of resections		135
Operative deaths	2	
Patients dead with associated cancer	4	
Patients lost to follow-up	8	
Patients convalescing	1	
Patients currently followed		120
Patients with periodic distress		3
Patients symptom-free and rehabilitated		117
Rehabilitation rate		97.5 per cent

deaths occurred in the entire group for whom resections were performed, a mortality rate of 1.5 per cent.

Sigmoidovesicle and/or sigmoidouterine fistulas were found in 17 men and 4 women. In 10 cases primary resections and partial cystectomies without colostomies were performed and 11 patients were operated on in stages. Although no deaths occurred in either group the postoperative complications in the group with single-stage procedures were significantly more marked than in the multiple-stage group. For this reason we consider the latter procedure preferable.

In this series 117 patients are free of symptoms and completely rehabilitated, a rehabilitation rate of 97.5 per cent.

Diverticulosis becomes more apparent in the fifth to the seventh decades with a corresponding tendency for the disease process to become accentuated. Patients in these age groups withstand poorly the ravages of repeated septic or hemorrhagic attacks with their complications, multiple operations and prolonged morbidity. We therefore urge those who treat diverticulitis to continue to broaden the scope for early surgical intervention and not to procrastinate from one attack to another.

The incidence of complications continues to be high but this should not be construed to represent the only indication for operation. While one-stage operations can and should be employed more frequently, even when certain complications are present, multiple-stage operations should be chosen when the patient's condition or the security of bowel continuity might be jeopardized with a one-stage procedure. One must select carefully the best operative procedure for each patient individually if good results are to be obtained.

Radical mastectomy with arterial regional chemotherapy

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Although the standard radical mastectomy as introduced by Halsted[10, 11] and Meyer[17] has been generally accepted as the most effective means of treatment for early carcinoma of the mammary gland, numerous other approaches have been made in an attempt to improve the end results. Superradical procedures added to the standard radical mastectomy have been investigated. It was Halsted,[10, 11] in 1898, who extended the range of dissection into the neck to attack the supraclavicular nodes. Later, Wangensteen[23] re-explored this approach, but, as Halsted had found, the results were unimpressive. W. S. Handley and his son have studied another avenue of metastatic spread[12, 13] and have shown the significance of the anterior mediastinal lymphatic chain and its surgical approach. Recently Urban[22] has reported 300 cases of en bloc resection of the internal mammary lymphatic chain in which the 5-year survival appeared to be 10 per cent greater than with the standard radical mastectomy. Considerable enthusiasm has been shown for the technique of simple mastectomy followed by radiation therapy as championed by McWhirter.[16] Haagensen's[9, 15] triple biopsy technique represents an attempt to establish the operability of breast lesions and individualize therapy.

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Management of widespread metastatic disease with ablative or additive hormone therapy has proved to be of some palliative value. Alterations of hormones have not been curative and their value in prophylaxis of metastases is not entirely clear.

In considering more extensive operations than that represented by classic radical mastectomy, it must be remembered that carcinoma of the breast may have already spread to areas that cannot be controlled by any local surgical procedure. Furthermore, operations of a more radical nature may be accompanied by increased morbidity, mortality rate, and disability. The number of viable tumor cells entering into the blood stream during the operation may be appreciable, and is in no way lessened by extending the operation. There is a growing belief that surgery with or without radiation therapy has approached its limit in the control of breast cancer and further major improvements in the results cannot be anticipated with these modalities. While it is true that the ultimate solution to breast cancer will probably be chemical or hormone management, there may well be an intermediate period when operation combined with chemotherapy will give improved results.[8]

In an attempt to enhance the effect of chemotherapeutic agents on a localized tumor, a method of administration of the chemical via the arterial route has been developed by 2 of the authors.[3, 4] Other in-

vestigators[1, 2, 7, 14, 20] have noted encouraging clinical results with this modality of treatment.

Our previous studies[3-5] have shown that (1) a higher concentration of chemotherapeutic agents, such as nitrogen mustard, can be delivered arterially into a localized tumor than by the usual intravenous route and (2) by occlusion of the corresponding venous return increased local effect of the chemical delivered arterially can be observed and this effect is roughly proportional to the time the venous occlusion is maintained.

The application of arterial regional chemotherapy (ARC) in combination with a radical operation on the breast might achieve several beneficial effects. First, while the artery chosen for injection is isolated, it is possible to ligate the accompanying veins early in the operation to reduce the possibility of tumor dissemination by this route. Second, malignant cells which might become dislodged during the operation are destroyed or seriously damaged through the ubiquitous effect of the high concentration of the chemical in the operative field. Third, malignant cells that have spread into neighboring tissue which ordinarily would not be encompassed by operation, but which is supplied by the artery, may be destroyed or severely damaged by the chemical.

MATERIAL

Twenty-one women with carcinoma of the breast treated with radical mastectomy and arterial regional chemotherapy from May, 1956, through June, 1959, are included in this preliminary study. The age range was 36 to 72 with an average of 54.8 years.

Fifteen of the 21 lesions were considered advanced. Skin retraction was noted in 11 patients, subcutaneous spread in 2, metastasis to the subcutaneous tissue near the midline in 1, and Paget's disease of the nipple with a subareolar tumor mass in 1. Four lesions were considered to be moderately advanced which measured over 4 cm. in diameter but showed no skin involvement. The remaining 2 were early lesions under 3.0 cm. in diameter with no involvement of the skin. The outlook in these cases was considered favorable.

The internal mammary artery was utilized on 4 occasions, the lateral thoracic artery on 8 occasions, the pectoral branch of the thoracicoacromial artery on 6 occasions, the subclavian artery on 2 occasions, and both the internal mammary and lateral thoracic artery on 1 occasion. In general, the artery supplying the segment of the breast containing the tumor was used.

The amount of nitrogen mustard* instilled into the selected arteries varied between 5 and 20 mg. In the case in which both the internal mammary and the lateral thoracic arteries were catheterized, 7 mg. of nitrogen mustard was administered into each artery. The nitrogen mustard was dissolved in saline to a concentration of 1 mg. per 1 ml. There was 1 case in which 20 mg. of nitrogen mustard was administered into the subclavian artery. Several of the patients with breast lesions of the medial hemisphere underwent internal mammary node dissection after the intra-arterial chemotherapy. Under these circumstances the entire lymphatic chain and the internal mammary artery and vein were removed en bloc from the first to the fourth intercostal space.[24]

ANATOMY

The internal mammary artery is the major source of blood to the breast (Fig. 1). The branches from the internal mammary artery via its anterior perforating arteries in the upper 4 intercostal spaces run forward, piercing the intercostal muscles and the pectoralis major, and reach the breast from behind. The second important arterial supply is the lateral thoracic artery which arises from the axillary artery along the lateral border of the pectoralis minor and descends along the lateral thoracic wall to supply the lateral half of the breast. The third artery of significance is the pectoral branch of the thoracicoacromial artery. These 3 major

*Mustargen hydrochloride (methyl bis(β-chloroethyl)-amine hydrochloride).

arteries, the internal mammary, the lateral thoracic, and the pectoral branch of the thoracicoacromial, in this order, supply not only the entire breast, but also the surrounding areas which ordinarily are not encompassed by radical surgical procedures. The venous drainage follows closely the arterial supply.

In an attempt to better understand the extent of the intra-arterial chemotherapy, we have studied the blood supply to the breast by administering fluorescein* at the time of the chemotherapy. Immediately after the injection of 5 ml. of fluorescein, upon illumination of the region with an ultra violet light, a spectacular bright yellow color manifesting the skin distribution of the artery was noted (Fig. 2).

TECHNIQUE

Selection of the internal mammary artery. After the usual mastectomy incision encompassing the breast was outlined, an incision was made within this outline 5 cm. long overlying the second rib cartilage as close to the sternum as the originally planned incision permitted. No supplemental incision was necessary (Fig. 3). The incision was immediately carried down to the chest wall and the second costal cartilage identified and dissected free. After placement of a small curved hemostat under this cartilage, the costochondral junction was divided with the scalpel. Then the intercostal muscle of the first and second intercostal spaces was divided. The cartilage was then elevated and deflected toward the sternum by fracture at the sternochondral junction. This brought the internal mammary artery and vein, together with the lymphatic chain, into view. The vein and the proximal end of the artery were ligated. A small incision was made in the arterial wall and a No. 18 or 20 polyethylene catheter inserted into the distal end of the artery for a distance of approximately 1 cm. and tied in place. Next, fluorescein

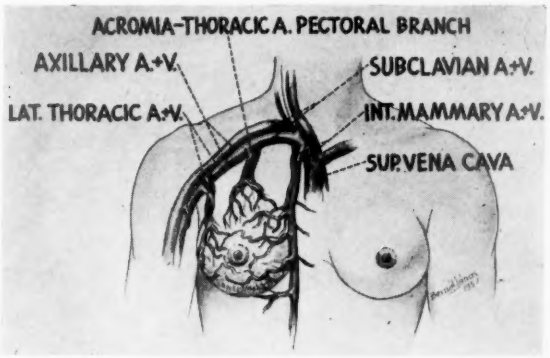

Fig. 1. Blood supply of the breast.

Fig. 2. Fluorescence after internal mammary artery injection.

was administered to outline the distribution of blood via this artery. After satisfactory demonstration of the blood supply was obtained, 5 to 10 mg. of nitrogen mustard was slowly injected. The catheter was removed and the artery ligated. The costal cartilage was sutured back into position with a single chromic suture. The radical mastectomy followed by the traditional Halsted method.

*Obtained as Fluorescite (5 per cent fluorescein with sodium bicarbonate), from the C. F. Kirk Co., New York, N. Y.

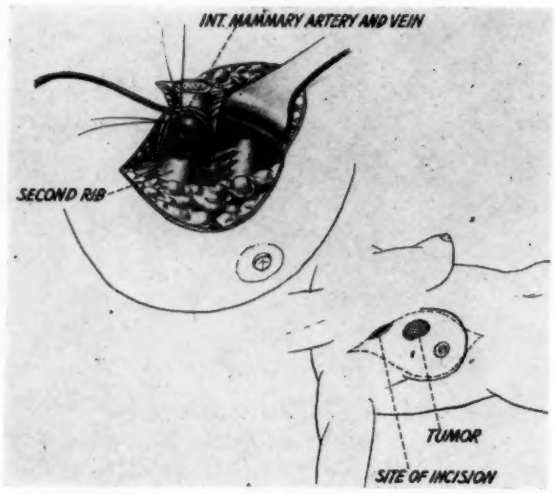

Fig. 3. Arterial regional chemotherapy technique.

The arterial regional chemotherapy was accomplished in approximately 15 minutes, and when the internal mammary artery had been ligated during the initial phase of the operation, the lost time could be partially regained during the ensuing operation because of the markedly reduced blood supply to the region.

Selection of the lateral thoracic and the pectoral branch of the thoracicoacromial artery. In the case of the selection of the lateral thoracic or the pectoral branch of the thoracicoacromial artery, the appropriate portion of the incision was made overlying these arteries. The pectoralis major was split or retracted as the occasion indicated, and the artery and vein were identified and treated as the internal mammary vessels had been.

Selection of the subclavian artery. In 2 instances the subclavian artery was utilized for chemotherapy of the breast. This was indicated when it was desirable to reach a wider area than usual with the chemical. The subclavian artery was approached just below the clavicle. Care was taken to occlude temporarily the artery below the ramification of the lateral thoracic to prevent the chemical from entering the arm. Because the subclavian artery was not sacrificed, a simple needle injection was carried out for the chemical administration. Venous occlusion of the subclavian vein was maintained for 5 minutes and then released.

RESULTS AND DISCUSSION

The 21 patients were followed for from 4 to 37 months. At present there are 7 patients living without evidence of local or distant recurrence. Three patients are living with recurrent disease. Of the latter, 2 have had local tumor in the skin which appeared from 4 to 25 months after therapy. Among the 10 patients who are known to be dead, 1 died with primary carcinoma of the ovary and 1 died after a gastrectomy for a benign gastric ulcer. The remaining 8 patients died directly from extensive metastatic disease. One patient is lost to follow-up at 4 months and is considered to be dead. Five of the 7 patients who are living and well without evidence of local or distant metastases had negative axillary and internal mammary nodes.

Complications consisted chiefly of sloughing of the superficial layer of the skin. Ten patients had moderate to mild skin necrosis (Fig. 4). Except in 1 case, all of the skin sloughs healed without skin grafting. The effect of intra-arterial nitrogen mustard therapy on the skin which did not slough consisted of erythema, edema, and hyperpigmentation. The histologic studies have been thoroughly discussed elsewhere.[21] There were no systemic effects. In our series there

Fig. 4. Skin effects of arterially administered nitrogen mustard. Note area of demarcation which will develop into a superficial slough.

was no bone-marrow depression or drop in the peripheral white blood cell count. The other formed elements of the blood, namely, the red cells and the platelets, showed no apparent changes after the application of this type of therapy.

Recently Creech[6] has introduced a new method of intra-arterial chemotherapy in which the extracorporeal heart-lung pump-oxygenator is utilized. Certain regions, such as the extremities and pelvis, are well suited for this type of experimental approach, but in lesions of the breast, where the blood supply is quite diversified, this type of regional therapy is not easily carried out. Moreover, the procedure requires a skilled team and a complicated apparatus which at present is not generally available.

Intravenous chemotherapy as an adjunct to surgical procedures has been investigated by Cole and his group[8, 19] and is being given a wide trial throughout the nation.[18] This combined approach has not been in use long enough to allow any definite conclusions to be drawn as yet, but statistics which are available suggest that there may be a place for chemotherapy utilized in this fashion. Arterial regional chemotherapy (ARC) is not time-consuming and the complications are minimal if the dose is kept to 10 mg. of nitrogen mustard or less. In arteries the size of the internal mammary and lateral thoracic, the dosage should never exceed 10 mg. ARC would appear to be superior to the usual intravenous chemotherapy as it permits the chemical to be delivered to the region in higher dosage and greater concentration.

SUMMARY

A method of arterial regional chemotherapy (ARC) in conjunction with radical mastectomy is described. Twenty-one patients have been treated in this fashion. There were no operative deaths. ARC is a practical technical adjunct to operations on the breast. No conclusions can be drawn concerning long-range benefits, but the method appears to have sufficient promise to warrant further studies.

REFERENCES

1. Barberio, J. R., Berry, N., Bateman, J., Cromer, J. K., and Klopp, C. T.: Combined administration of aureomycin and nitrogen mustard. II. Effects of the intra-arterial administration on human cancer, Cancer **6:** 280, 1953.
2. Barberio, J. R., Klopp, C. T., Ayres, W. W., and Gross, H. A.: Effect of intra-arterial administration of nitrogen mustard, Cancer **4:** 1341, 1951.
3. Bierman, H. R., Byron, R. L., Jr., Miller, R., and Shimkin, M. B.: Effects of intra-arterial administration of nitrogen mustard, Am. J. Med. **8:** 535, 1950.
4. Bierman, H. R., Byron, R. L., Jr., Miller, R., and Shimkin, M. B.: Intra-arterial catheterization in man, Bull. Univ. California M. Center **1:** 84, 1949.
5. Byron, R. L., Jr., Singh, B. P., Bierman, H. R., and Kelly, K. H.: Intra-aortic nitrogen mustard therapy in advanced pelvic malignancies, SURGERY **45:** 634, 1959.
6. Creech, O., Jr., Krementz, E. T., Ryan, R. F., and Winblad, J. N.: Chemotherapy of cancer: regional perfusion utilizing an extracorporeal circuit, Ann. Surg. **148:** 616, 1958.
7. Cromer, J. K., Bateman, J. C., Berry, G. N., Kennelly, J. M., Klopp, C. T., and Platt, L. I.: Use of intra-arterial nitrogen mustard therapy in the treatment of cervical and vaginal cancer, Am. J. Obst. & Gynec. **63:** 538, 1952.
8. Cruz, E. P., McDonald, G. O., and Cole, W. H.: Prophylactic treatment of cancer, SURGERY **40:** 291, 1956.
9. Haagensen, C. D., and Obeid, S. J.: Biopsy of the apex of the axilla in carcinoma of the breast, Ann. Surg. **149:** 149, 1959.
10. Halsted, W. S.: A clinical and histological study of certain adenocarcinoma of the breast, Ann. Surg. **28:** 557, 1898.
11. Halsted, W. S.: The results of radical operations for the cure of cancer of the breast, Ann. Surg. **46:** 1, 1907.
12. Handley, R. S., and Thackray, A. C.: The internal mammary lymph chain in carcinoma of the breast, Lancet **2:** 276, 1949.
13. Handley, W. S.: Parasternal invasion of the thorax in breast cancer and its suppression by the use of radium tubes as an operative precaution, Surg. Gynec. & Obst. **45:** 721, 1927.
14. Klopp, C. T., Alford, T. C., Bateman, J., Berry, G. N., and Winship, T.: Fractionated intra-arterial cancer chemotherapy with methyl bis amine hydrochloride; preliminary report, Ann. Surg. **132:** 811, 1950.
15. McDonald, J. J., Haagensen, C. D., and

Stout, A. P.: Metastasis from mammary carcinoma to the supraclavicular and internal mammary lymph nodes, SURGERY **34:** 521, 1953.

16. McWhirter, R.: Simple mastectomy and radiotherapy in the treatment of breast cancer. Brit. J. Radiol. **28:** 128, 1955.
17. Meyer, W.: An improved method of the radical operation for carcinoma of breast, New York M. Rec. **46:** 746, 1894.
18. Moore, G. E., and Kondo, T.: Study of adjuvant cancer chemotherapy by model experiments, SURGERY **44:** 199, 1958.
19. Mrazek, R. G., Economou, S. G., McDonald, G. O., Harridge, W. H., Slaughter, D. P., and Cole, W. H.: Chemotherapy and prophylactic measures in the treatment of cancer, S. Clin. North America **39:** 41, 1959.
20. Sullivan, R. D., Jones, R., Jr., Schnabel, T. G., and Shorey, J. M.: The treatment of human cancer with intra-arterial nitrogen mustard utilizing a simplified catheter technique, Cancer **6:** 121, 1953.
21. Sullivan, R. D., Mescon, H., and Jones, R., Jr.: The effect of intra-arterial nitrogen mustard therapy on human skin, Cancer **6:** 288, 1953.
22. Urban, J. A.: Clinical experience and results of excision of the internal mammary lymph node chain in primary operable breast cancer, Cancer **12:** 14, 1959.
23. Wangensteen, O. H., Lewis, F. J., and Stuart, W. A.: The extended or super-radical mastectomy for carcinoma of the breast, S. Clin. North America **36:** 1051, 1956.
24. Yonemoto, R. H., and Byron, R. L., Jr.: Radical mastectomy with conservative anterior mediastinal node dissection: The "trapdoor approach," SURGERY **47:** 908, 1960.

Perforation of a metastatic carcinoma of the jejunum after cancer chemotherapy

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Cancer chemotherapy has a potential for altering current concepts concerning malignant diseases. With the discovery and use of newer therapeutic agents, it is quite conceivable that greater success is to be expected from the application of such drugs.

The tumoricidal effect of chemical substances may be a mixed blessing, however, under certain circumstances. One such situation may be when a tumor comprises a portion of the wall of a hollow viscus. The tumor, in this instance, serves a useful function while at the same time contributes to the death of the patient. Should generalized, rapid tumor necrosis occur, the wall of the hollow viscus of which the tumor is a part will also undergo necrosis. This could lead at once to the catastrophic development of a perforated hollow viscus.

The purpose of this report is to present an unusual instance of metastasis of a bronchogenic carcinoma. An occult metastasis occurred to the jejunum which was not discovered until a perforation through this tumor occurred after chemotherapy.

CASE REPORT

The patient was a 36-year-old white male Air Force sergeant who, in December, 1958, noted swelling of the hands, feet, and left knee. Evaluation at an Air Force Hospital lead to the discovery of a mass in the left hemithorax which was compatible with a bronchogenic carcinoma. Results of bronchoscopic examination and scalene node biopsy were reported as negative.

On Jan. 16, 1959, a left thoracotomy was performed which disclosed a "nonresectable bronchogenic carcinoma." Only a biopsy of the tumor was performed, and this showed anaplastic carcinoma. Radiation therapy was administered after operation. The patient was discharged from the Air Force and transferred to the Coral Gables Veterans Administration Hospital on Feb. 22, 1959. He remained under observation for 3 days and was then discharged. During the next few months he was asymptomatic and gained 18 pounds in weight. In June, however, he began to experience severe interscapular pain and was readmitted to the Veterans Hospital on June 19, 1959. A chest roentgenogram showed a left hilar mass, but roentgenograms of the thoracic spine made on admission and several times thereafter were negative. Clubbing of the fingers and toes at this was quite marked. A course of cyclophosphamide† therapy was started on June 21, 1959. This consisted of the intravenous administration of 600 mg. daily for 5 days. Hematologically, the chemotherapy was well tolerated. After this treatment, pain in the back diminished, but the patient began to experience hemoptysis and pedal edema. The 5-day course of cyclophosphamide therapy was repeated beginning July 20, 1959. On July 25, 1959, 1 day after the completion of the second course of chemotherapy, severe abdominal pain developed. The pain was persistent and not completely controlled by narcotics. When seen by the surgical service the patient presented marked abdominal tenderness and rigidity. Roentgenographic examination showed the presence of free peritoneal air. On July 26, an operation was performed. The preoperative diagnosis had been "perforated peptic ulcer." At operation a generalized peritonitis was

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†Cytoxan, manufactured by Mead Johnson & Company, Evansville, Ind.

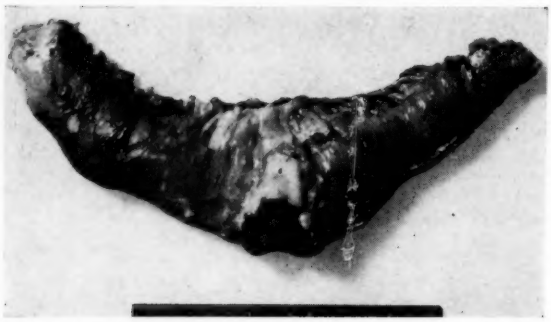

Fig. 1. Gross specimen of segment of jejunum containing metastatic tumor with perforation through the tumor.

present. The gastroduodenal area appeared normal. Further exploration disclosed a perforation through a tumor of the jejunum approximately 2 feet distal to the ligament of Treitz. The tumor measured 7 by 5 by 3 cm. and was located on the antimesenteric border of the intestine. The perforation was approximately 0.5 cm. in diameter. No other intra-abdominal metastases were noted. An 18-inch segment of jejunum containing the tumor was resected and an end-to-end anastomosis performed. The gross specimen is shown in Fig. 1. The patient's recovery from the operation was satisfactory except for the development of a superficial wound infection. His general condition continued to deteriorate slowly and he died on Sept. 16, 1959, approximately 7 weeks after the bowel perforation.

On postmortem examination a tumor was found arising from the bronchus of the left upper lobe with extension involving the chest wall, pericardium, and heart. Numerous metastases were found in the liver, gallbladder, small intestine, colon, kidneys, adrenal glands, thyroid gland, and mediastinal, retroperitoneal, and mesenteric lymph nodes. Histologic examination of the bronchial tumor showed a small-cell anaplastic carcinoma and these sections were identical to the sections obtained from the small bowel perforation (Figs. 2 and 3).

DISCUSSION

Metastatic tumors of the small intestine, while uncommon, are not a rare clinical entity. De Castro[4] reported 26 cases and was able to find reports of an additional 25 cases in 1955. Since 1955 we have found the reports of 6 other instances,[1-3, 5-7] for a total of 57. The case of our patient is not presented merely as another example of this type of tumor spread. The patient's clinical course demonstrates the first reported instance of the perforation of a small bowel metastasis after chemotherapy. De Castro[4] reported perforations of metastatic small bowel tumors but none of them had occurred after chemotherapy.

Whether the perforation in this instance

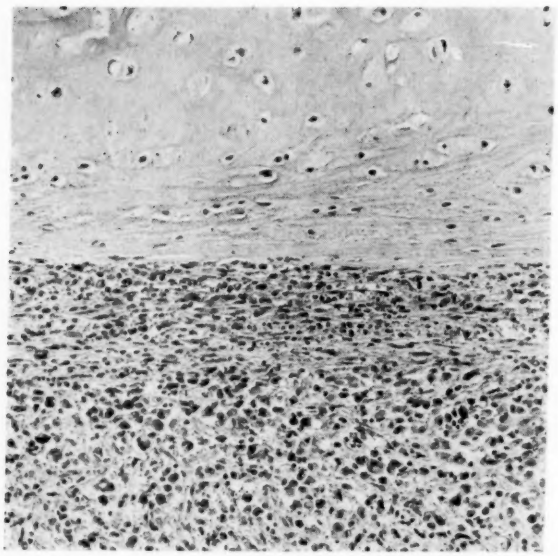

Fig. 2. Photomicrograph of anaplastic carcinoma in bronchus. (×75; reduced ⅔.)

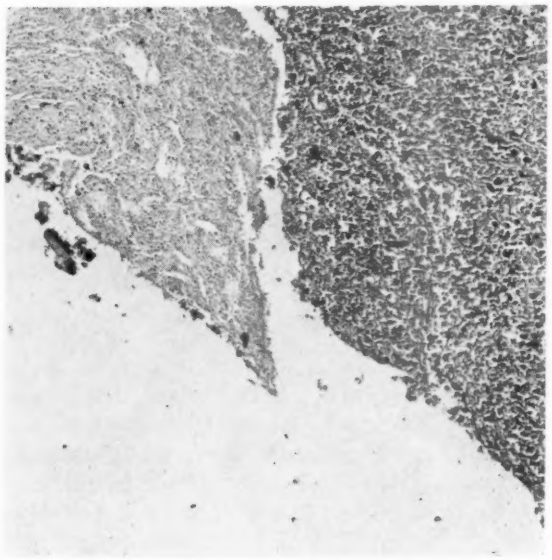

Fig. 3. Photomicrograph of anaplastic carcinoma in jejunum. An area of necrosis is seen at the left. (×75; reduced ⅔.)

was "spontaneous" or due to the chemotherapy is, of course, conjectural. In support of the latter contention is the fact that the patient enjoyed a salutory response after the first course of chemotherapy and that the perforation occurred immediately after the second course of therapy.

It is not the purpose of this report to indict or to question the use of chemicals in the treatment of malignant diseases. Their future effectiveness will probably be of great benefit to patients stricken with malignant tumors. However, with the development of more effective chemotherapeutic drugs, physicians must be alerted to the possibility of occasional undesirable results with this treatment. One such result may be the perforation of a hollow viscus after chemotherapy when a part of the wall of the viscus is actually composed of tumor. It would be wise, therefore, for physicians working in institutions or in situations in which cancer chemotherapy is applied to consider this possibility. It is likely that, as the chemotherapy drugs increase in effectiveness, this complication may be encountered with increasing frequency.

SUMMARY

A case of perforation of a metastatic tumor of the small bowel after cancer chemotherapy is presented. The relationship of the chemotherapy to this occurrence and the possible resulting implications are discussed.

REFERENCES

1. Case Records of the Massachusetts General Hospital: Case 44171, New England J. Med. **258:** 846, 1958.
2. Case, T. C.: Ileoileal intussusception of a solitary metastatic tumor of the ileum, New York J. Med. **58:** 2280, 1958.
3. Conte, M., Conte-Marti, M., Baledent, P., and Frelot, C.: Le cancer metastatizue de l'intestin grele, Semaine hôp. Paris **35:** 461, 1959.
4. De Castro, C. A., Dockerty, M. B., and Mayo, C. W.: Metastatic tumors of the small intestines, Surg. Gynec. & Obst. **105:** 159, 1957.
5. De Jode, L. R.: A case of metastatic carcinoma of the ileocaecal valve presenting as a primary growth, Brit. J. Surg. **46.** 505, 1959.
6. Gatch, W. D.: A melanoma, apparently primary in a breast; its single known metastasis in the small bowel, A. M. A. Arch. Surg. **73:** 226, 1956.
7. Von Capeller, D., and Cummins, A. J.: Solitary duodenal metastasis eight years after radiation of carcinoma of the cervix, Gastroenterology **33:** 655, 1957.

The clinical and pathologic aspects of pleural mesotheliomas

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For years the contention that primary malignant tumors arise from pleural lining cells has been challenged by the prevailing belief that primary lung cancer is responsible for the pleural growths.

After analysis of 5 cases, with the use of the whole-lung section to depict the distinctive pleural growth of primary tumors arising from mesothelial lining cells, we are able to question the prevailing concept. Fahr[4] has stated, "in its total aspect, it [the macroscopic picture of pleural mesotheliomas] is much more typical than the microscopic." The cytologic components of pleural mesotheliomas have been delineated, with particular reference to the histogenesis of the parent cell.

HISTORICAL BACKGROUND

The first reference to tumors of the pleura was made by Joseph Lientaud in about 1767. Engelbach,[2] in 1891, was the first to raise the question of whether such tumors arose from the endothelium of the lymph vessels or from the cells lining the serous surfaces. Before this, Wagner's[14] concept of lymph channels filled with tumor cells governed the majority of the theories concerning the origin of these tumors. Some[3, 11, 15] have denied the existence of primary mesotheliomas as such. Ewing,[3] along with Robertson,[11] approved the term "endothelioma," but said that only sarcomas can be classified as primary malignant tumors of the pleura. As early as 1924 Robertson[11] said, "Rarely have the students of any other subject in Medical Science proposed such a bewildering array of contradictory theories."

The incidence of these tumors is about 1.1 cases per 1,000 autopsies, according to a series of 45,000 autopsies reported by Saccone and Coblenz.[12] Hochberg[7] collected 60,042 autopsies, with an incidence of 0.07 per cent.

No instance appears to have been recorded in which a metastatic tumor involved the pleura as completely as do mesotheliomas. McCaughey[9] pointed out that in 305 cases of primary lung parenchymal carcinomas, there were only 5 cases in which the pleura was extensively infiltrated. Of 608 carcinomas from other sites, only one, a carcinoma of the thyroid, simulated the appearance of a pleural neoplasm. The great majority of secondary neoplasms in the pleura form discrete nodules, but do not possess the more regular sheetlike character ascribed to most pleural mesotheliomas.

HISTOGENESIS

Hertwig and Hertwig,[6] in 1881, showed that the lining of the pleuropericardial cavity apparently develops from the coelomic epithelium which is split off from the mesoderm. Coelomic epithelium gives rise to definite glandular structures as well as to serosal cells. Malignant mesothelial cells may revert to a more immature state, and they do possess the potency to form alveolar structures. Miller and Wynn[10] were the first to advance the opinion that a neoplasm of

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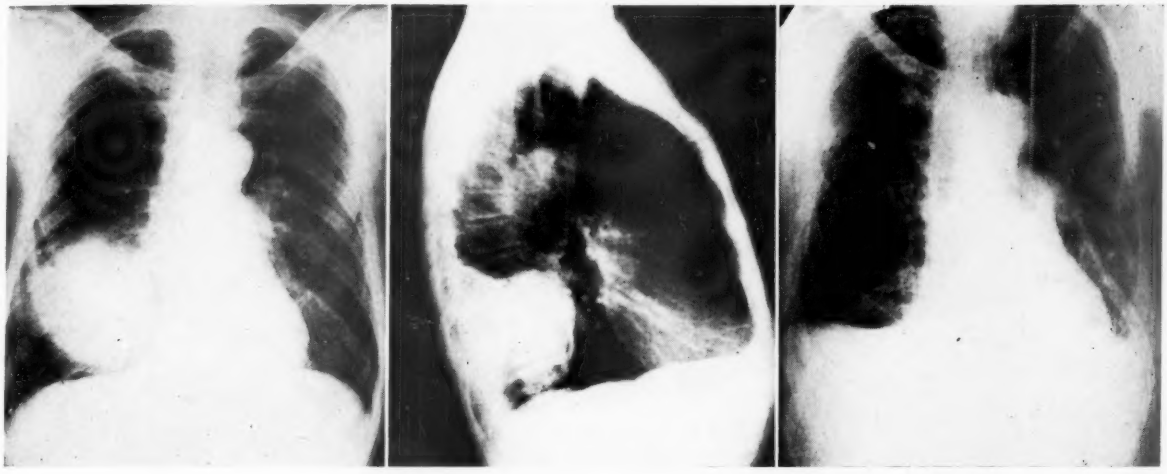
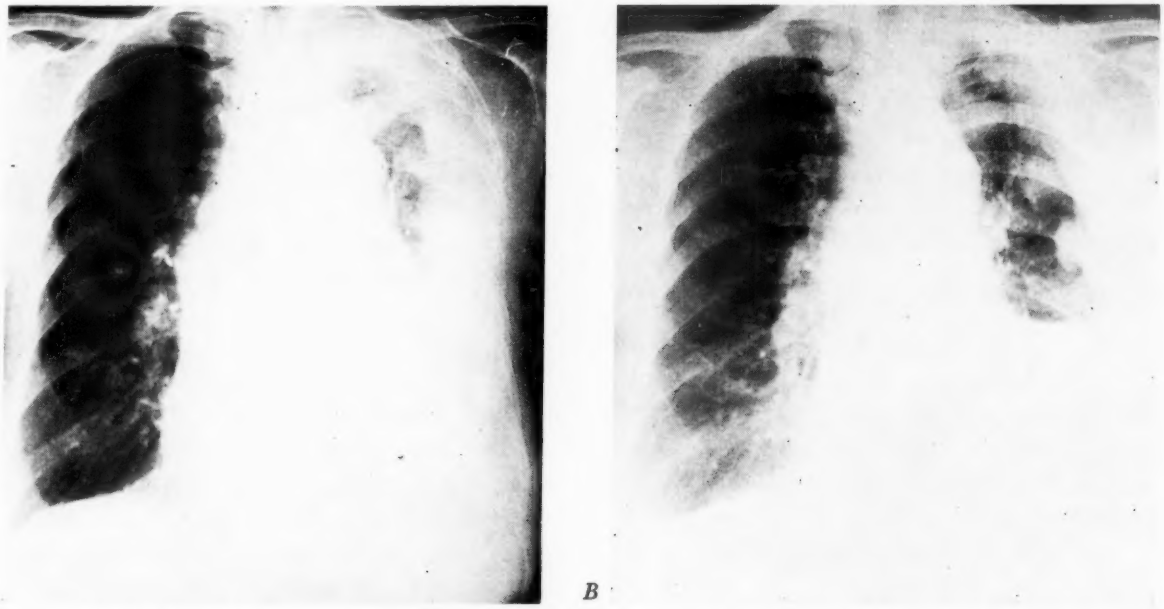
the pleura showed both fibroblastic and epithelial characteristics because of the embryonic relation of the cells to the mesoderm.

Maximow,[8] using tissue culture techniques, showed that the mesothelium or covering cells of the serosal surfaces can form cells of a fibroblastic appearance. Stout and Himadi[13] also demonstrated in tissue cultures that mesothelial cells can



A *B* *C*

Fig. 1. Case 1. *A* and *B,* Preoperative roentgenograms of pleural mesothelioma involving periphery of right lower lobe and adjacent posterior parietal pleura. Note the 2 large nodular lesions. *C,* The picture within 1 week of operation (right lower lobectomy and pleurectomy).



A *B*

Fig. 2. Case 5. *A,* Pleural mesothelioma of left lung and pleura before thoracentesis. *B,* After thoracentesis. Note the nodular densities of the pleura and lung. Pleural fluid examination showed malignant cells.

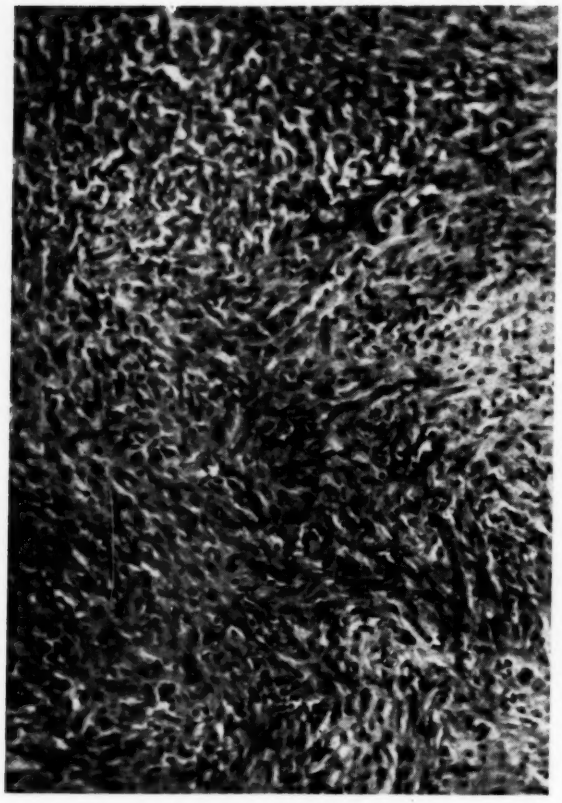


Fig. 3. Case 1. A representative section which shows a fibrosarcomatous pattern. (×120; reduced 1/6.)

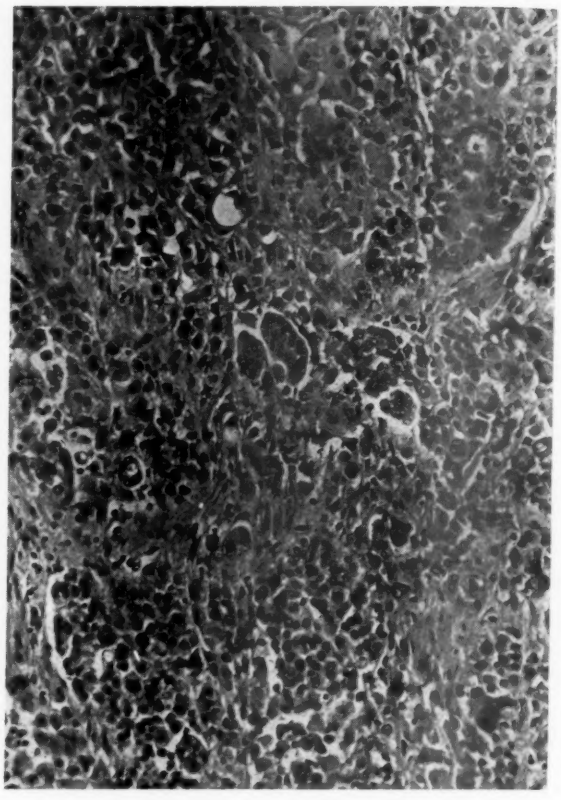


Fig. 4. Case 5. A representative section which shows a glandular pattern. (×120; reduced 1/6.)

develop into solid, tubular, or glandular structures of epithelial character as well as into mesenchymal tissues with fibroblasts, collagen, and reticulum fibers.

Wilder's silver stain demonstrated a marked paucity of reticulum in these lesions, which supports the belief that these cells, which are of mesodermal origin, do not exhibit reticuloendothelial characteristics.

CLINICAL ASPECTS

The age incidence of our 5 patients with primary malignant pleural mesotheliomas ranged between 52 and 72 years. This coincides with the ages in most cases reported in the literature, although Bogardus[1] reported a case in a 3-year-old white girl.

The most common complaints of our patients were shortness of breath and chest pain. Pleural effusions were noted to be present in 4 of 5 cases. This sign is much more prevalent in patients with this neoplasm than in those with primary parenchymal neoplasms of the lung.

Chronic inflammatory lesions of the lung (such as tuberculosis or actinomycosis), malignancy elsewhere (such as ovary or breast) with metastasis to the lung and pleura, the so-called "collagen diseases" of the lung, pleura, and/or pericardium, and even simple heart failure may cause chest pain and pleural effusion and the roentgenographic findings may not be characteristic. The problem of differential diagnosis then becomes a most perplexing one. Exploratory thoracotomy may be necessary.

The roentgenographic findings were not specifically characteristic, but were highly suggestive. Pleural effusion and the peripheral nodular pulmonary lesions were easily discernible in many instances (Figs. 1 and 2).

These neoplasms are highly malignant lesions, as exemplified by the fact that all 5

of our patients were dead within 10 months of the onset of symptoms. Godwin,[5] in his series which included 10 malignant neoplasms arising from the pleura, had only 3 patients which survived for more than 12 months after the onset of symptoms. When these neoplasms are first detected, they are usually nonresectable (4 of our 5 cases). At postmortem examination, however, the lesions are not found to be widely disseminated beyond the confines of the chest (Table I).

HISTOPATHOLOGY

The lesions in Cases 1, 2, and 4 showed a dominantly fibrosarcomatous pattern (Fig. 3), with diffuse, dense proliferation of plump, spindle-shaped cells which had pleomorphic, slightly hyperchromatic nuclei. In contrast, the lesions in Case 5 showed a predominantly glandular pattern (Fig. 4), the cells were markedly pleomorphic and bizarre, and the nuclei showed marked increase in size and definite hyperchromation. Only the lesion in Case 3 showed a predominantly mixed pattern.

MACROSECTION OF THE LUNG

The large lung section, which was originally introduced by Gough, was used to great advantage to capture the gross picture of pleural mesotheliomas. The thick encasement of the lung by the tumor, with growth all along the interlobar fissure, is well illustrated by this method (Fig. 5). The method provides a morphologic "map" which allows

Table I. *Summary of 5 cases*

Case No.	*Age at death*	*Sex*	*Known duration of illness (mo.)*	*Autopsy*	*Site of primary lesion*	*Important clinical symptoms and signs*	*Treatment*	*Extension and metastatic spread*
1 (E. M.)	58	White male	9	No	Right pleura	Chest pain, cough, weight loss, pleural effusion, mass on roentgenogram	Pleurectomy, lobectomy, x-ray therapy	On re-examination by roentgenogram; right pleural cavity, right middle and upper lobes
2 (E. B.)	52	White male	2	Yes	Left pleura	Chest pain, weight loss, dyspnea, pleural effusion, mass on roentgenogram		Whole left lung, parietal pericardium, left diaphragmatic pleura
3 (W. C.)	66	White male	10	Yes	Right pleura	Chest pain, dyspnea, anorexia, weight loss, mass on roentgenogram		Right lung, right interlobar fissure, liver
4 (L. P.)	63	White male	5	Yes	Left pleura	Chest pain, dyspnea, pleural effusion, mass on roentgenogram	Decortication	Left lung, right parietal and visceral pleura, right lung, pericardium, heart, liver, gall bladder, spleen, gastrointestinal tract, adrenal glands, kidneys, urinary bladder, peritoneum, omentum
5 (J. O.)	72	White female	3	Yes	Left pleura	Fever, cough, dyspnea, pleural effusion, mass on roentgenogram	Intravenous nitrogen mustard	Left lung, left diaphragmatic pleura, anterior mediastinum

Fig. 5. Case 5. Whole lung section which shows encasement of the lung by the pleural mesothelioma, with extension along the interlobar fissure.

one to see the unusual and peculiar pathway of these lesions.

CONCLUSION

The pleomorphic appearance of these tumors, indicated by the stromal and glandular elements, suggests a pleuropotential line of development. The macroscopic whole lung section, as illustrated particularly in Case 5, delineates the typical picture of a primary pleural mesothelioma. This technique is thought to lend credence to the theory that the pleura is the site of primary origin of these tumors.

SUMMARY

Five cases of primary mesotheliomas of the pleura have been presented, with introduction of the macroscopic whole lung section to better illustrate these lesions. As far as we know, it is the first time this technique has been used to outline this particular lesion.

The cases which have been presented were all similar in that all 5 patients were dead within 1 year of the onset of symptoms, and that all patients were between the sixth and eighth decade of life.

Chest pain, pleural effusion, and confluent peripheral nodular pulmonary lesions are highly suggestive of a primary pleural mesothelioma. Exploratory thoracotomy is necessary to confirm this diagnosis.

We are grateful for being allowed to study and present the cases of Drs. David Johnson, St. Louis, Mo. (Case 2); John Pfaff, Great Falls, Mont. (Case 4); and James Nakada, St. Louis, Mo. (Case 5).

REFERENCES

1. Bogardus, G. N., Knudtson, K. P., and Mills, W. H.: Pleural mesothelioma, Am. Rev. Tuberc. **71:** 280, 1955.
2. Engelbach: Ein Beitrag zur differential Diagnose pleuritischer Exudate und Neubildungen der Pleura in Anschluss daren ein Fall von Endothelcarcinom der Pleura, Inaug.-Diss., Freiburg, 1891.
3. Ewing, J.: Neoplastic diseases, Philadelphia, 1922, W. B. Saunders Company, ed. 2, p. 319.
4. Fahr, T.: Zur Frage des sog. Pleuraendothelioms, Virchow's Arch. f. path. Anat. **295:** 502, 1935.
5. Godwin, M. C.: Diffuse mesotheliomas, Cancer **10:** 298, 1957.
6. Hertwig, O., and Hertwig, R.: Die coelomtheorie: Versuch einer Erkiärung des mittieren keimblattes, Ztschr. f. Naturwissensch. **8:** 1, 1881.
7. Hochberg, L. A.: Endothelioma (mesothelioma) of the pleura, Am. Rev. Tuberc. **63:** 150, 1943.
8. Maximow, A.: Über das Mesothel (Deckzellen der serösen Häute) und die Zellen der serösen Exsudate, Untersuchungen an entzündetem Gewebe und an Gewebskulturen, Arch. f. exper. Zellforsch. **4:** 1, 1927.
9. McCaughey, W. T. E.: Primary tumors of the pleura, J. Path. & Bact. **76:** 517, 1958.
10. Miller, J., and Wynn, W. H.: A malignant tumor arising from the endothelium of the peritoneum and producing a mucoid ascitic fluid, J. Path. & Bact. **12:** 267, 1908.
11. Robertson, H. E.: Endothelioma of the pleura, J. Cancer Res. **8:** 317, 1924.
12. Saccone, A., and Coblenz, A.: Endotheliomas of the pleura, Am. J. Clin. Path. **13:** 186, 1943.

13. Stout, A. P., and Himadi, G. W.: Solitary (localized) mesothelioma of the pleura, Ann. Surg. **133:** 50, 1951.
14. Wagner, E.: Das tuberkelähnliche Lymphadenom. (Der cytogene oder reticulirte Tuberkel.) Arch. f. Heilk. **11:** 495, 1870.
15. Willis, R. A.: A metastatic deposit of bronchial carcinoma in a hydrocele misdiagnosed, "endothelioma," J. Path. & Bact. **47:** 35, 1939.

Pediatric surgery

Conducted by MARK M. RAVITCH, M.D.

Nonmechanical conditions simulating obstructive lesions of the intestinal tract in the newborn infant

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The great importance of prompt recognition of mechanical obstruction in the neonatal period has been repeatedly stressed. Physicians who are the first to see the newborn infant should be alert in the detection of such conditions so that they may be brought quickly to the attention of pediatric surgeons who have become highly skilled in their correction. As a result of our aggressiveness in such situations we have encountered several conditions which mimic intestinal obstruction so closely that differentiation is exceedingly difficult. Under such circumstances the pediatric surgeon will often decide to err on the side of safety and perform an exploratory operation on these infants. It is our purpose in this report to call attention to 3 recent experiences encountered by us at The Cook County Children's Hospital.

CASE REPORTS

Case 1. Baby W., born prematurely to a heavily sedated mother, weighed 3 pounds, 3 ounces, and required resuscitation to initiate respirations. During the first few hours of life she was tachypneic and cyanotic. A roentgenogram demonstrated atelectasis of the right lung. Air in the gastrointestinal tract was limited to the stomach. The baby became tense and hyperreflexic while her respiratory difficulties diminished. Although oral feedings were withheld she had a coffee-ground emesis on the third day. Bile was not grossly evident. Repeated x-ray examination with Lipiodol instilled into the stomach via gastric catheter showed that neither air nor contrast media left the stomach in 72 hours (Fig. 1). It was recognized that she had central nervous system damage, but a pyloric obstruction was considered highly probable in view of the roentgenographic findings. When the infant was 80 hours of age celiotomy failed to reveal any evidence of mechanical obstruction. She survived for 8 days after operation, during which time the symptoms of obstruction persisted. At autopsy a patent gastrointestinal tract was confirmed and severe encephalomalacia was demonstrated.

Case 2. Baby Mc. was a 7-pound boy born at home to a heroin addict. At 2 days of age he was admitted to this hospital with symptoms of respiratory distress, irritability, and hyperactive reflexes. Shortly after admission he vomited bile and initial roentgenograms of the abdomen showed air in the stomach with only small flecks of air noted in the distal intestinal tract (Fig. 2, *A*). A roentgenogram taken the next day with air injected through a catheter in the stomach showed similar findings (Fig. 2, *B*). A presumptive diagnosis of partial duodenal obstruction was made but at operation no obstructing mechanism could be found. Routine postoperative care resulted in improvement of the infant's condition and 2 days later he was able to retain his formula. He was discharged home on the twenty-first postoperative day in good condition.

Case 3. Baby G. was seen by the surgical consultants at the age of 3 days because of vomiting which was reported to contain no bile. Her birth weight was 5 pounds. Upright roentgenograms of the chest and abdomen were considered normal as were contrast-media studies of the upper gastro-

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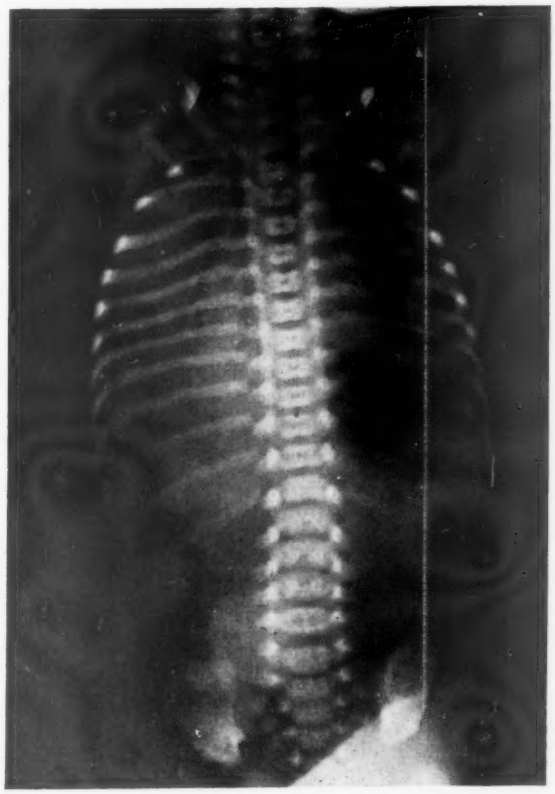
intestinal tract. Medical management was continued and retained oral feedings were supplemented with appropriate fluids given parenterally. In spite of this regimen, the infant lost weight to 4 pounds, 4 ounces, on the thirteenth day of age. Serum electrolyte determinations showed a progressive fall in the chloride level despite what were thought to be adequate intravenous doses of normal saline solution. At 2 weeks of age the serum sodium and potassium levels were normal, but the serum chloride level was 69 mEq. per liter. Two days later a surgical consultant felt a small tumor in the right hypochondrium. Peristaltic waves from left to right were prominent across the upper abdomen. This was confirmed by other observers and a repeat roentgenogram with barium was thought to confirm the impression of congenital hypertrophic pyloric stenosis (Fig. 3). At exploratory operation there was a normal pylorus and no evidence of obstruction of the stomach or duodenum. The seromuscular layers of the pylorus were incised, permitting easy passage of a gastrostomy tube into the duodenum. Vomiting persisted and the clinical course as well as the continued low serum chloride levels finally prompted the diagnosis of adrenal insufficiency. Immediate improvement was noted when desoxycorticosterone acetate (DOCA) therapy was started along with the intravenous administration of sodium chloride. This therapy was continued for 14 days, after which the DOCA was gradually reduced and finally discontinued on the eighteenth postoperative day. At the time of her discharge from the hospital, 1 week after DOCA therapy had been discontinued, the infant had regained her birth weight and vomiting did not recur.

DISCUSSION

The cardinal symptoms of mechanical obstruction of the intestinal tract in the newborn infant are vomiting and abdominal distention. If the obstruction is high, distention may be absent or limited to the upper abdomen. Vomiting is persistent and usually forceful. The presence of bile in the emesis depends upon the relationship of the obstructive lesion to the ampulla of Vater. In the presence of such symptoms, x-ray examination helps to confirm the diagnosis of obstruction. On an upright film of the chest and abdomen the gastrointestinal gas pattern will usually delineate the site of the obstruction. The use of contrast media may be necessary in doubtful cases, and even then a false impression may be obtained as in our cases.

Smith[11] describes the gastrointestinal tract of the newborn infant as showing completely developed digestive and absorptive surfaces as well as cellular elements similar to those



A

B

Fig. 1. Baby W. was operated upon on the fourth day of life for suggestive pyloric obstruction. No lesion was found at operation. Autopsy 8 days later showed no gastrointestinal lesion, but a severe encephalomalacia. *A,* Upright film taken at 6 hours shows atelectasis of the right lung and no gas beyond the stomach. *B,* Lipiodol study, oblique view, taken when infant was 72 hours of age, shows complete retention of contrast material.

of the adult. The suppportive musculature, however, is deficient, as are the elastic fibers of the submucosa. As a result of this, distention occurs readily in the neonate. There is also much variation in the gastric size, depending on the amount of air and liquid swallowed. Most investigators, according to Smith,[12] are in general agreement with regard to the passage of air along the gastrointestinal tract in normal newborn infants. Air is visualized in the stomach often within 5 minutes after birth and in the jejunum within 15 minutes. The cecum shows evidence of air after 2 hours.[8] Because this process is so consistent and rapid, roentgenographic evidence of interference with air filling helps greatly in the diagnosis of obstruction along the gastrointestinal tract in the newborn infant.[16]

The preoperative diagnosis of pyloric obstruction in our first patient was justified on the basis of the known, but rare, existence of this lesion in the neonate. Touroff[15] and more recently Benson and Coury[2] have reported successful treatment of pyloric obstructions in the newborn infant. The infant in Case 1 was prematurely born and showed evidence of cerebral damage. The explanation for the failure of this baby's stomach to empty may have been the absence or destruction of the motility centers in the cerebral cortex.[1]

The second baby exhibited classic signs of narcotic poisoning in the newborn infant. The characteristic shrill cry, hyperirritability, and coarse tremors have been emphasized by many who have described this syndrome.[4, 10, 13] Vomiting was a prominent symptom stressed in most reports. Ten of 22 infants mentioned by Cobrinik and associates[3] vomited or fed poorly. Biliary vomiting was not described.

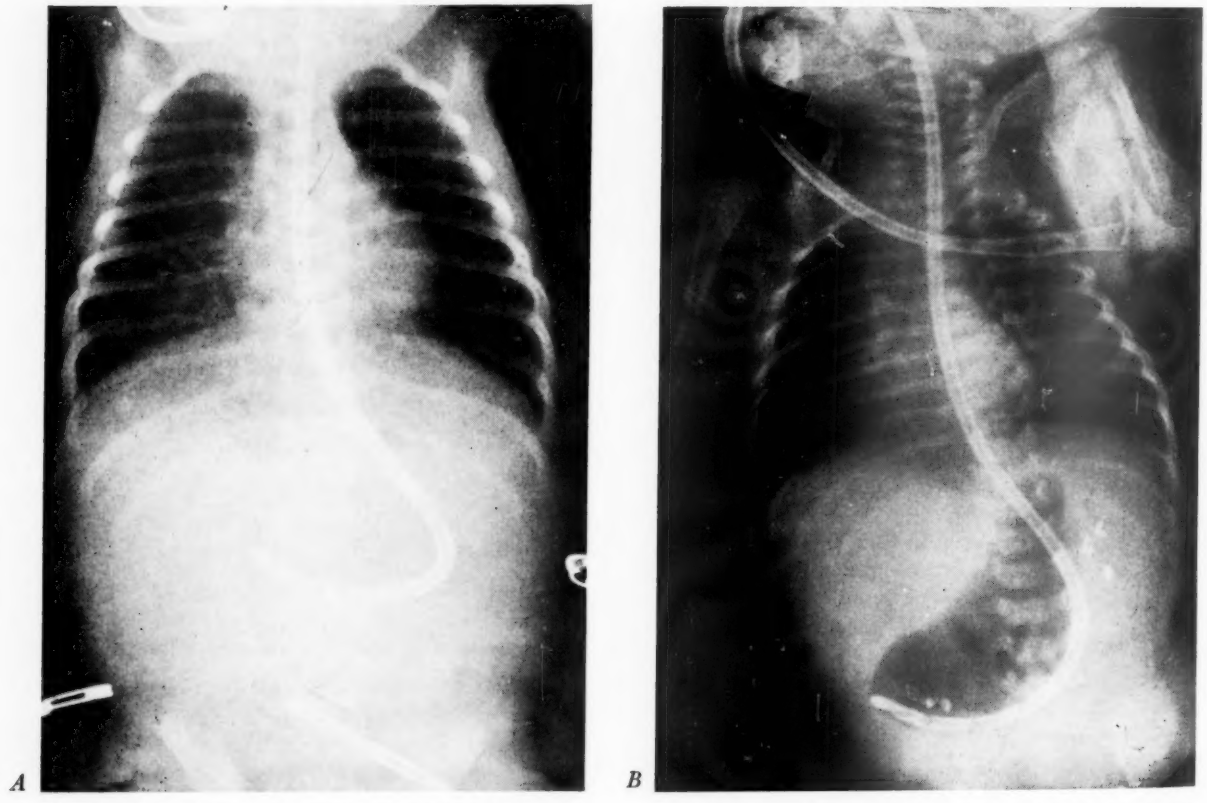


Fig. 2. Baby Mc. was born of a heroin addict and operated upon on the third day of life with a presumptive diagnosis of partial duodenal obstruction. *A*, Plain film taken at 48 hours shows no air in the gastrointestinal tract. *B*, At 72 hours, air insufflated into the stomach does not pass into the duodenum and only an occasional gas shadow is seen elsewhere in the abdomen.

The association of pyloric stenosis and adrenal cortical hyperplasia was pointed out by Pirie[7] in 1919. Since then reports have stressed the similarity between adrenal cortical insufficiency and pyloric stenosis.[5, 9] Weens and Golden[17] reported 2 such cases. "Most infants with abnormal adrenocortical function present as (1) those in whom the deficient secretion of hormone regulating water and salt metabolism induces severe temporary or permanent electrolyte disturbances, (2) those having adrenal hyperplasia with excessive liberation of adrenal androgens, girls in this group manifest pseudohermaphroditism, and (3) those having a combination of the first two groups." When a salt-losing syndrome is a prominent feature, marked dehydration and severe vomiting may occur. The latter may be projectile and so dominate the picture that a diagnosis of hypertrophic pyloric stenosis may be made.[6] It may be very difficult to differentiate the 2 conditions, even on x-ray examination, and many such patients have been operated upon. In our third patient x-ray examination suggested hypertrophic pyloric stenosis but exploratory operation failed to show an obstructive lesion. Absence of a palpable mass in the right hypochondrium in a baby with suspected pyloric stenosis should alert one to the possibility of adrenal insufficiency. Severely low serum chloride levels may be found in infants with pyloric stenosis alone, as emphasized by Terezis and colleagues.[14] The two conditions may coexist, and may require both operative and endocrine treatment. Twenty-four hour collection of urine from a baby for steroid determination may be difficult. Empirical treatment with DOAC and normal saline in babies with suspected adrenal insufficiency might be the best course to follow.

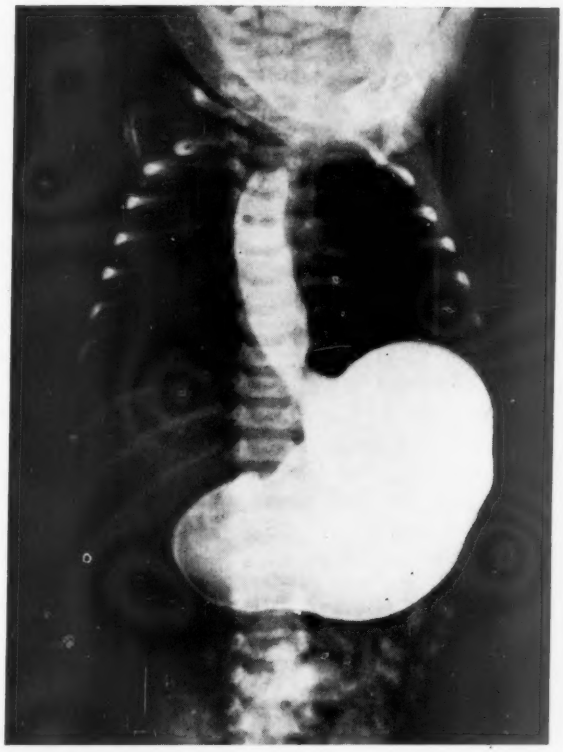


Fig. 3. Baby G. was operated upon at 2 weeks of age for persistent projectile vomiting and gastric peristaltic waves. No lesion was found and persistently low serum chloride levels led to treatment with desoxycorticosterone acetate. All symptoms were relieved. Roentgenogram taken after a barium swallow shows prolonged retention in the greatly dilated stomach, interpreted as compatible with pyloric stenosis.

SUMMARY

Three cases of suspected high gastrointestinal obstruction in newborn infants are described. The failure to demonstrate a gross anatomic obstructing lesion at operation led to investigation of other possibilities. These proved to be (1) gross cerebral defect, (2) narcotic poisoning in the infant of an addict, and (3) adrenocortical insufficiency. The significance of an accurate history of the prenatal maternal status, the birth history, and neonatal laboratory findings in the infant are discussed.

REFERENCES

1. Babkin, B. P., and Kite, W. C., Jr.: Central and reflex regulation of motility of the pyloric antrum, J. Neurophysiol. **13:** 321, 1950.
2. Benson, C. D., and Coury, J. J.: Congenital intrinsic obstruction of the stomach and duodenum in the newborn, A. M. A. Arch. Surg. **62:** 856, 1951.
3. Cobrinik, R. W., Hood, R. T., Jr., Chusid, E., and Slobody, L. B.: The effects of maternal narcotic addiction on the newborn infant, A. M. A. J. Dis. Child. **92:** 504, 1956.
4. Krause, S. O., Murray, P. M., Halmer, J. B., and Burch, R. E.: Heroin addiction among

pregnant women and their newborn, Am. J. Obst. & Gynec. **75:** 754, 1958.

5. Morec, G. J.: Hypertrophic pyloric stenosis associated with adrenal insufficiency, J. Pediat. **42:** 359, 1953.
6. Munck, W.: Pseudo-pyloric stenosis in diffuse hyperplasia of the adrenal cortex in infants, Acta path. et microbiol. scandinav. **91:** 24, 1951.
7. Pirie, G. R.: A study of hyper-adrenalism: its influence in producing congenital pyloric hypertrophy and subsequent stenosis, Lancet **97:** 513, 1919.
8. Podolsky, M. L., and Jester, A. W.: The distribution of air in the intestinal tract of infants during the first twelve hours as determined by serial roentgenograms, J. Pediat. **45:** 633, 1954.
9. Rosenberg, P.: Temporary adrenal insufficiency with associated pyloric stenosis, J. Pediat. **48:** 63, 1956.
10. Schneck, H.: Narcotic withdrawal symptoms in the newborn infant resulting from maternal addiction, J. Pediat. **52:** 584, 1958.
11. Smith, C. A.: Physiology of the newborn infant, Springfield, 1959, Charles C Thomas, Publisher, ed. 3, pp. 230-231.
12. Smith, C. A.:[11] p. 235.
13. Steg, N.: Narcotic withdrawal reactions in the newborn, A. M. A. J. Dis. Child. **94:** 286, 1957.
14. Terezis, N. L., Sieber, W. K., and Kiesewetter, W. B.: The prognostic significance of electrolyte and weight studies in congenital hypertrophic pyloric stenosis, Surg. Gynec. & Obst. **109:** 482, 1959.
15. Touroff, A. S. W., and Sussman, R. M.: Congenital prepyloric membranous obstruction in a premature infant, SURGERY **8:** 739, 1940.
16. Wasch, M., and Marck, A.: The radiographic appearance of the gastrointestinal tract during the first day of life, J. Pediat. **32:** 479, 1948.
17. Weens, H. S., and Golden, A.: Adrenal cortical insufficiency in infants simulating high intestinal obstruction, Am. J. Roentgenol. **74:** 213, 1935.

Long-term results of rectosigmoidectomy for Hirschsprung's disease, with a note on Duhamel's operation

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After Hirschsprung's[8] first description of the disease to which his name has been attached, 60 years elapsed before the essential problems concerning the etiology and treatment of the disease were solved.[1, 4, 14, 15] Swenson's[14] operation was soon recognized as the first and only adequate treatment for aganglionic megacolon and became universally adopted. Since 1948 a considerable number of patients with Hirschsprung's disease all over the world have been cured by this operation.

In recent years, long-term results of Swenson's operation have been assessed in a number of follow-up studies.[6, 7, 12, 16] Excellent results with a low mortality rate and few complications were found by Swenson[12] in the largest of the series reported. In reports from the Hospital for Sick Children, London,[16] Presbyterian Hospital, New York,[7] and Kinderspital, Zürich,[6] attention has been focused on a fairly high incidence of early and late postoperative complications.

This report is an additional contribution to the evaluation of long-term results of rectosigmoidectomy for Hirschsprung's disease. The present series consists of 69 patients treated consecutively from 1948 to 1959. There were 55 boys and 14 girls in this series. Preliminary colostomies were performed in 22 cases and omitted in the remaining 47 cases. One total colectomy and one hemicolectomy are included. The length of the aganglionic segment ranged from 6 to 35 cm. in the remaining cases. Ages at operation and operative mortality rate are listed in Table I. Three deaths occurred, 1 at operation (cardiac arrest) and 2 the early postoperative period (1 fulminant tracheobronchitis and 1 leak from the anastomosis with diffuse peritonitis). A fourth patient made an uneventful recovery after rectosigmoidectomy but died from a sudden aspiration on the ward after colostomy closure 6 weeks later.

Early postoperative complications occurred in 22 patients as listed in Table II. Deaths and complications in patients with and without a preliminary colostomy are shown in Table III. One patient died and 3 patients had residual symptoms from their complications. All of the remaining 18 patients were cured with or without additional treatment.

The 65 patients who survived rectosigmoidectomy have all been followed for from 1 to 12 years after operation, 52 of them for from 5 to 12 years. They are all alive at the end of this follow-up period. General health, growth, and development have been excellent in all patients.

Thirty-five patients have been relieved of all symptoms and signs of disease. In 8 of these patients, however, temporary disturbances of bowel function persisted for up to 7 years after operation (Table IV).

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Table I. *Ages at operation and operative mortality rate*

Age	*No. of patients*	*Deaths*
11 days	1	—
6-12 mo.	23	3
1-2 yr.	10	—
2-5 yr.	9	1
5-14 yr.	26	—
Totals	69	4

The remaining 30 patients still have some kind of residual disturbance of bowel and/or bladder function 1 to 12 years after operation (Table V). Constipation is defined as lack or rare occurrence of spontaneous movements. Laxatives and enemas are given regularly and with good results. The patients with diarrhea all have soft stools, mostly once or twice and sometimes 3 to 4 times daily. Repeated attacks of acute gastroenteritis have been of common occurrence in these patients. Diarrhea is mostly combined with fecal incontinence. The degree of fecal and urinary incontinence is slight in the majority of cases. The figures given in parentheses in Table V denote the number of patients with pronounced symptoms.

DISCUSSION

The relatively high incidence of early and late complications found in this series should not obscure the fact that more than half of the patients have been completely relieved of their disease and that the majority of patients with residual disturbances are only slightly inconvenienced and are otherwise perfectly healthy. Only 4 patients died. Compared to the fate of patients with Hirschsprung's disease before the introduction of rectosigmoidectomy, these are excellent results indeed.

The operative mortality rate is somewhat higher (about 6 per cent) than in the series reported earlier (1.5 to 4 per cent), as listed in Table VI. There seems to be no con-

Table II. *Early complications*

Complication	*No.*	*Treatment*	*No.*	*Result*
Incomplete resection	6	Secondary operation	3	Cured
		No further treatment	3	1 normal function, 1 residual constipation, 1 diarrhea, anal incontinence (mongolism)
Leak from anastomosis	5	Colostomy and/or incision	4	3 cured, 1 anal incontinence
		Conservative	1	Dead (peritonitis)
Stricture of anastomosis	7	Dilatations	7	Cured
Wound rupture	2	Resuture	2	Cured
Intestinal obstruction	1	Lysis of adhesions	1	Cured
Urinary retention	1	Catheter	1	Cured
Totals	22			18 cured, 3 had residual symptoms, 1 dead

Table III. *Deaths and complications in patients with and without preliminary colostomies*

Operation	*No.*	*No. of deaths*	*Incomplete resection*	*Leaks*	*Strictures*
With colostomy	22	1	6	0	2
Without colostomy	47	3	0	5	5
Totals	69	4	6	5	7

Table IV. *Temporary functional disturbances after rectosigmoidectomy*

Disturbance	*No.*	*Postoperative duration (yr.)*	*Additional treatment*	*Duration of cure (yr.)*
Constipation	1	4	Sphincterotomy	1
Diarrhea	4	1-7	1 sphincterotomy	1-8
Anal incontinence	4	2-5	—	4-8

clusive difference between patients treated with and without preliminary colostomy (Table III). No preoperative and no late postoperative deaths occurred in the present series. The over-all mortality rate is nearly the same as in all of the other series (6 to 8 per cent). The predominance of infants in mortality statistics (Table I) is in agreement with earlier reviews.

The list of early complications (Table II) includes 6 incomplete resections. The incidence of this technical error is related to the objections of our pathologists to the assessment of the adequacy of resection by frozen sections. The proximal resections have been performed according to roentgenologic and gross pathologic criteria. As seen in Table III, all of the incomplete resections were performed in patients with colostomy. Some of these patients were very ill at the time colostomy was performed. A formal laparotomy in order to determine and mark the level of the transitional cone was sometimes considered inadvisable, and the surgeon was thus deprived of an otherwise excellent landmark. As stressed in earlier reports, dilatation of the distal colon may disappear after colostomy. Identification of the transitional cone at subsequent rectosigmoidectomy may thus become very difficult. This experience is fully borne out in the present series.

Secondary resections were performed in 3 patients with satisfactory end results. In a fourth patient bowel function has been normal for 10 years after incomplete resection. One patient has residual constipation which is easily managed and 1 is a mongoloid with diarrhea and fecal incontinence.

Incomplete resections are mentioned in earlier follow-up studies but their incidence is not reported. Frozen sections from the level of proximal resection offer the only safe method for assessment of adequacy of resection.

Gross leak from the anastomosis occurred in 5 cases. A circumscribed perforation was found dorsally in the midline of the anastomosis in all of these cases. One patient died of diffuse peritonitis 5 days after operation. The remaining 4 patients developed pelvic abscesses, which were drained and the bowel decompressed by transverse colostomies. Three of these patients made uneventful recoveries. In the fourth case a perineal fistula developed. Division of the fistula after long-standing suppuration resulted in persistent anal incontinence.

All of the leaks occurred in patients without preliminary colostomies (Table III). As suggested in an earlier report,[5] inadequacy of vascular supply near the mesenteric attachment to the bowel favors the development of necrosis in this area. Postoperative bowel paralysis coupled with swelling of the anastomosis and/or sphincter spasm may re-

Table V. *Persistent functional disturbances after rectosigmoidectomy*

Disturbance	*No.*	*Comments*
Constipation	5	1 incomplete resection, 1 mental retardation
Diarrhea	18	14 with anal incontinence
Anal incontinence	22 (8*)	1 sphincter division after pelvic abscess and fistula, 3 mental retardation
Urinary incontinence	16 (7*)	12 with anal incontinence

*Figures in parentheses denote number of patients with pronounced symptoms.

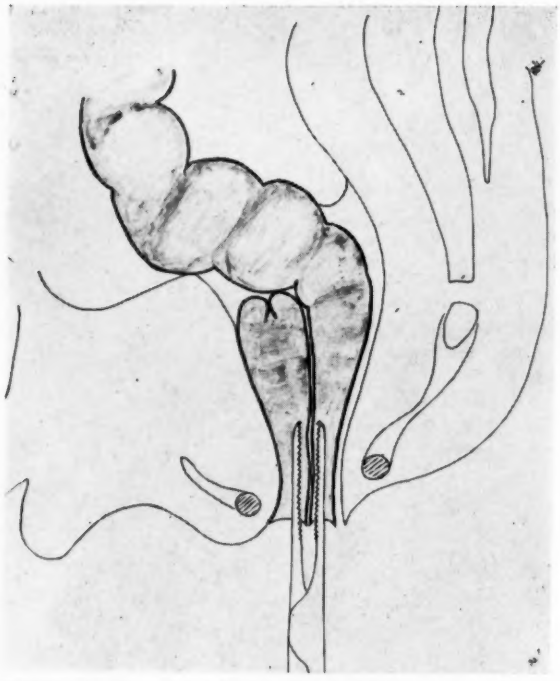

Fig. 1. The principle of Duhamel's operation.

sult in distention of the distal bowel. This added impediment to circulation is sometimes enough to cause necrosis. A preliminary colostomy prevents excessive dilatation of the distal bowel and should thus decrease the risk of necrosis. The risks, however, are not fully preventable. Leaks have also been reported to occur in patients with colostomy.[16]

The incidence of postoperative strictures of the anastomosis was similar to that found in other series (Table VI). All responded to treatment with dilatations and none of the patients had residual symptoms.

The other complications listed in Table II were few and of no importance for the final outcome.

The most distressing finding in this follow-up study is the high incidence of residual disturbances of bowel and bladder function (Tables IV and V). Diarrhea and constipation are probably symptoms of the same functional disturbance. Fecal and urinary incontinence are of common occurrence in this group of patients. Repeated complete studies in a number of these patients, including cystometric studies, voiding urethrocystograms, and defecation studies by barium enema, have failed to reveal any underlying pathologic factors. In particular, signs of neurogenic bladder disturbance were absent in all of the cases investigated.

Over a number of years, some of these patients ultimately have been relieved of their symptoms (Table IV). In the majority of cases, however, the disturbance has persisted (Table V). On the assumption that tightness of the remaining aganglionic segment of the internal sphincter could be related to the complication,[3, 9, 12, 13] sphincterotomies have been performed in 3 cases. Two of the patients (Table IV) were completely relieved from constipation and diarrhea, respectively. The third patient still suffers from constipation 1 year after sphincterotomy.

The incidence of residual constipation and/or diarrhea in previous follow-up studies shows considerable variation, ranging from 2.5 per cent to 30 per cent (Table VI). The same applies to fecal incontinence (0 to 27 per cent) and urinary incontinence (0 to 15 per cent).

Table VI. *Synopsis of follow-up studies after rectosigmoidectomy*

		Deaths			*Early complications*			*Late complications*			
										Incontinence	
Author	*No. of cases*	*Preoperative*	*Operative*	*Later*	*Incomplete resection*	*Leak*	*Stricture*	*Constipation*	*Diarrhea*	*Anal*	*Urinary*
Wyllie	152	—	6	3	3?	9	7	34	—	41	15
Swenson	200	—	6	7	—	5	9	1	4	0	0
Hiatt	67	—	1	4	—	—	—	—	4	9	—
Genton and Vontobel	32	6	1	1	—	—	4	8	2	7	5
Present series	69	0	4	0	6	5	7	5	18	22(8)	16(7)

In recent years attempts have been made to avoid the complications of rectosigmoidectomy by the use of modified operations. Anterior resections have again been advocated[9, 10] and rejected.[7, 12] The rationale of deliberately leaving a long aganglionic segment behind at operation is difficult to understand, considering the failures which occurred after inadvertently incomplete resections.

A somewhat new approach has been advanced by Duhamel[3]—a radical resection without pelvic dissection. The principle is to divide the bowel at the proximal end of the rectum, to close and leave the rectum in place, to resect the remaining portion of the aganglionic colon, and to bring normal colon through the retrorectal space down to the dorsal part of the anal opening, where a side-to-side anastomosis is created by application of 2 crushing clamps (Figs. 1 and 2).

According to Duhamel, his method offers several advantages: (1) no pelvic dissection and no danger of neurogenic disturbances, (2) no end-to-end anastomosis and no danger of leaks or strictures, (3) preservation of the anterior aganglionic part of rectum, which should warrant the start of defecation reflex, with the new posterior ganglionic part of the "rectal" wall providing for adequate motility, and (4) low operative risks, which would permit definitive operation in the very young infant.

In the last few years, Duhamel's procedure has gained increasing popularity on the European continent.[2, 6, 11] Grob[6] has recently modified the technique, applying the crushing clamps above the internal sphincter level in order to avoid anal incontinence.

During the last year we have tried the Duhamel procedure in 8 cases with satisfactory results. There were no deaths and no complications in this small series. We have been impressed by the ease with which the procedure is accomplished. This seems to make it particularly suited for use in the young infant. Because we are dealing with a congenital lesion, often requiring treatment of the neonate or young infant, the Duhamel procedure may prove to be an important contribution, provided that follow-up will confirm the expectations engendered by the primary results so far available.

Fig. 2. Barium enema study taken after Duhamel's operation.

SUMMARY

A follow-up study of 69 patients with Hirschsprung's disease, treated by rectosigmoidectomy from 1948 to 1959, is reported. Four patients died. Early postoperative complications were encountered in 22 cases. Temporary disturbances of bowel function occurred in 8 patients and persisted for from 1 to 7 years after operation. Remaining constipation or diarrhea, mostly combined with anal and urinary incontinence, was found in 30 patients. The majority of these patients had mild symptoms only and were otherwise perfectly healthy. Thirty five patients were relieved of all symptoms and signs of disease. Compared to the fate of patients with Hirschsprung's disease before introduction of rectosigmoidectomy, these results are considered to be excellent.

A new technical approach, recently advanced by Duhamel,[3] is described and has

been tried in 8 cases with satisfactory primary results. It is too early as yet to evaluate the potential advantages of the Duhamel procedure.

REFERENCES

1. Bodian, M., Stephens, F. D., and Ward, B. L. H.: Hirschsprung's disease and idiopathic megacolon, Lancet **1:** 6, 1949.
2. Duhamel, B.: A new operation for the treatment of Hirschsprung's disease, Arch. Dis. Child. **35:** 38, 1960.
3. Duhamel, B.: Technique chirurgicale infantile, Paris, 1957, Masson & Cie.
4. Ehrenpreis, Th.: Megacolon in the newborn, Acta chir. scandinav., vol. 94, supp. 112, 1946.
5. Ehrenpreis, Th.: Rectosigmoidectomy for Hirschsprung's disease, Acta chir. scandinav. **102:** 251, 1952.
6. Genton, N., and Vontobel, V.: Megacolon congenitum *in* Linneweh, F.: Die Prognose chronischer Erkrankungen, Berlin, 1960, Springer-Verlag.
7. Hiatt, R. B.: A further description of the pathologic physiology of congenital megacolon and the results of surgical treatment, Pediatrics **21:** 825, 1958.
8. Hirschsprung, H.: Stuhlträgheit Neugeborener in Folge von Dilatation und Hypertrophie des Colons, Jahrb. f. Kinderh. **27:** 1, 1888.
9. Rehbein, F., and Zimmermann, H.: Results with abdominal resection in Hirschsprung's disease, Arch. Dis. Child. **35:** 29, 1960.
10. State, D.: Surgical treatment for idiopathic congenital megacolon (Hirschsprung's disease), Surg. Gynec. & Obst. **95:** 201, 1952.
11. Sulamaa, M.: Personal communication, 1960.
12. Swenson, O.: Follow-up on 200 patients treated for Hirschsprung's disease during a ten-year period, Ann. Surg. **146:** 706, 1957.
13. Swenson, O.: Personal communication, 1960.
14. Swenson, O., and Bill, A.: Resection of rectum and rectosigmoid with preservation of the sphincter for benign spastic lesions producing megacolon, SURGERY **24:** 212, 1948.
15. Whitehouse, F. R., and Kernohan, J. W.: Myenteric plexus in congenital megacolon, Arch. Int. Med. **82:** 75, 1948.
16. Wyllie, G. G.: Treatment of Hirschsprung's disease by Swenson's operation, Lancet **1:** 850, 1957.